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METABOLIC CONTROL AND TREATMENT OF MALIGNANT GROWTH: TUMOR DEVELOPMENT, PATTERNS OF AMINO ACID IMBALANCE AND UKRAIN

NEFYODOV L.I.,[1] DOROSHENKO Y.M.,[2] SMIRNOV V.Y.,[2] UGLYANITSA K.N.,[3] NOWICKY W.[4]

1) Department of Biochemistry, Yanka Kupala State University of Grodno, Grodno, Belarus.
2) Central Scientific Laboratory, Grodno Medical School, Grodno, Belarus.
3) Department of Oncology, Grodno Medical School, Grodno, Belarus.
4) Ukrainian Anti-Cancer Institute, Vienna, Austria.

Summary: *In experimental (W-256, SM-1 and PC-1 tumors) and clinical (breast, bladder and prostate cancers) studies, the use of Ukrain was proven to be safe and highly effective against cancer, inhibiting protein synthesis in cancer cells, selectively accumulating in tumor tissue after a single intravenous administration and controlling cancer-induced metabolic imbalance. Ukrain inhibits metabolic processes in the tumor, causing metabolic disorders in cancer cells and affecting the transport and intermediate metabolism of amino acids involved in the processes of malignant growth. The cancerostatic action of Ukrain is achieved by controlling amino acid pool formation in the host organism and tumor, preventing the active transportation of amino acids into cancer cells and inhibiting some metabolic processes essential for the development of cancer.*

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Introduction

Development of a malignant tumor is accompanied by metabolic changes manifested both in the organism as a whole and in individual organs, tissues and cells, as well as in enzymatic reactions. These changes are primarily governed by the nature of the tumor, as a structure consisting of low differentiated cells displaying such special features as uncontrolled growth, metastatic spread, down-regulated apoptosis and necrosis (1).

In a cancer-bearing organism, various biochemical reactions are relatively reduced, amino acid metabolism enzyme reactions are of low rate and the regulatory mechanisms for maintaining metabolic balance are disturbed. Active malignant growth leads to competition for intermediate metabolic substrates

Address for correspondence: L.I. Nefyodov, Department of Biochemistry, Yanka Kupala State University of Grodno, ul. Ozheshko 22, 230023 Grodno, Belarus.
E-mail: l.nefyodov@grsu.by

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and energy between the tumor and the host organism (2). This competition is manifested in the ability of tumor cells to use host intermediates for energy generation and for their own growth. The tumor itself manifests high rates of various metabolic processes, in particular those requiring the production of considerable amounts of energy, and the substrate supply becomes a limiting factor in its growth (3, 4).

In a malignant tumor, the increased necessity of some amino acids induces and enhances their functional deficiency, manifesting as amino acid imbalance both in the tumor-bearing host and in the tumor itself. The extent of the resulting imbalance is probably determined by the amino acid transport activity into host and tumor tissues (5, 6). Competition between the host and the tumor considerably limits the potential for metabolic correction and treatment in cancer patients (7).

A further important factor determining the biochemical relationships between tumor bearer and tumor is the phase of tumor growth. Massive tumor cell necroses and accumulation of large amounts of underoxidized products induce changes in the redox potential and metabolic imbalance in host tissues (8).

The metabolic disorders described above are characteristic of cancer cells and are particularly aggravated by alterations in amino acid metabolism. Our interest in studying metabolic imbalance and patterns of free amino acid pool formation is based on a number of factors: in malignant growth, the deficiency of relatively and absolutely essential amino acids in patients with anorexia is accompanied by activation of endogenous protein and amino acid degradation, thus leading to a negative nitrogen balance (9); free amino acid absorption and transport are disturbed (7, 10); cell redox potential and relationships between anabolic and catabolic reactions in amino acid metabolism are changed (11); the toxic effect of tumor degradation products is blocked by directly interacting amino acids (cysteine, lysine) or their derivatives (glutathione), thus aggravating their deficiency (12); the relationships between metabolic pathways (glycolysis, gluconeogenesis) conjugated with intermediate amino acid metabolism and energy production reactions are altered (2, 13, 14); and some amino acids and their derivatives, *e.g.,* glutamine, are used as natural regulators of proliferation, differentiation and apoptosis in malignant cells (15, 16).

These factors confirm the importance of considering the mechanisms of amino acid imbalance in malignant growth for targeted metabolic therapy of cancer (17, 18).

Results and discussion

On the basis of existing published data and the results of our studies, we suggest that the biological activity of Ukrain against malignant growth is realized *via* several relatively independent mechanisms (19). Firstly, the cancerostatic effect of the agent is mediated by a decrease in activities essential for tumor growth, resulting in limited metabolic reactions, inhibition of angiogenesis, and enhancement of collagen production, with formation of a connective tissue capsule around the primary tumor that prevents its growth and the spread of metastases, as well as the activation of proteolysis and the degradation of structural elements of the cancer cells (Fig. 1). Secondly, there are the immune-modulating and immune-correcting effects of Ukrain and the activation of local immunity. Thirdly, there is the alleviation of metabolic imbalance, the ratios between the rates of endogenous compound chemical conversion along different pathways that generate and expend energy in normal cells by affecting the regulatory links in metabolic flows in the tumor-bearing organism (metabolic correction and therapy) (20) (Fig. 2).

Of principal importance is the application of Ukrain as a drug for the neoadjuvant therapy of cancer,

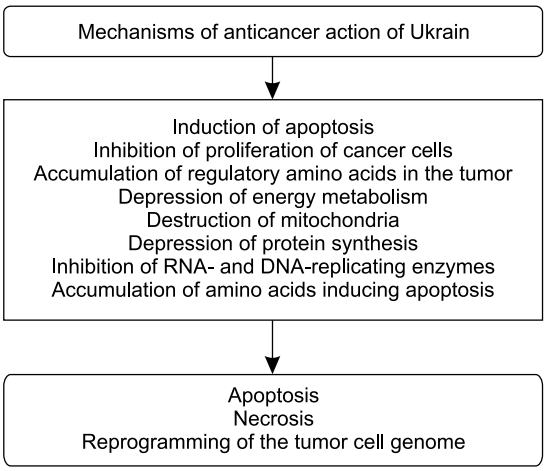

Fig. 1 Range of activities of Ukrain relevant to its anticancer effects.

which, in addition to the direct cytostatic and immune modulating effects of the substance, enables the implementation of the major principles of metabolic therapy: the targeting of the regulatory link in the metabolism responsible for general manifestations of metabolic imbalance, the goal of systemic and long-term action and the desirability of applying the drug in combination with other methods of anticancer therapy.

Therefore, along with the essential absence of side effects and time limits, sufficiently high specificity and the enhancement of other kinds of treatment, the application of the drug facilitates the achievement of the major goal of metabolic therapy: increasing the efficacy and decreasing the side effects and complications of conventional therapy (8, 21) (Fig. 1).

The specific effects of Ukrain when applied experimentally and clinically enabled us to conclude that, compared to other chemotherapeutic drugs, Ukrain has a bimodal (cytostatic/cytolytic and immunomodulating) mechanism of specific antitumoral effect, and that the metabolic effect of the drug can be realized on several levels (20) (Fig. 3).

In studying the pharmacokinetics, it was shown that compared to healthy animals, tumor bearers had a relatively selective accumulation of the drug in tumor tissue. The tissue-Ukrain binding was reversible, and the prolonged presence of the drug in the tumor-bearing organism was explained by a selective and

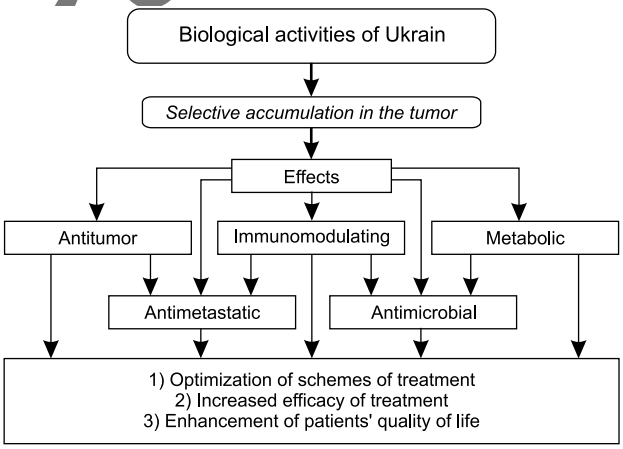

Fig. 2 Range of biological activities of Ukrain.

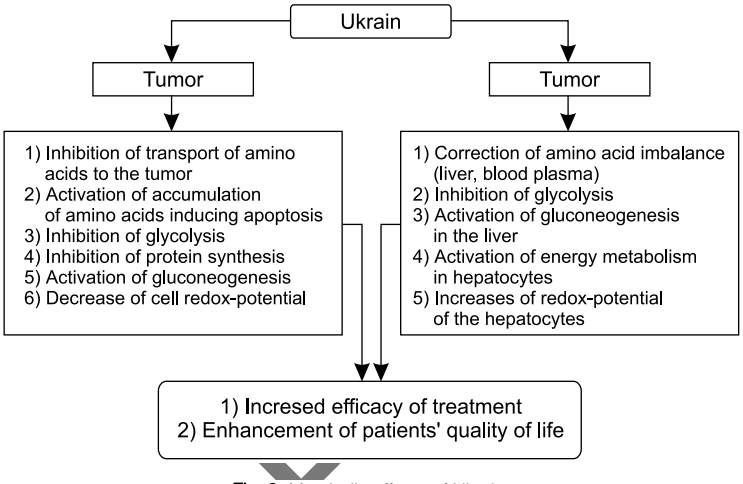

Fig. 3 Metabolic effects of Ukrain.

more rapid penetration into tumor tissue in comparison with normal tissue (22).

Such effect gives us grounds for believing that the specific antitumoral and metabolic effects found both experimentally and clinically are directly induced by Ukrain and related to its selective accumulation in tumor tissue (19).

In this situation, the unidirectional metabolic effects of Ukrain on free amino acids and their derivatives (reinforcement of the overall pool of glucogenic and thiol-containing amino acids) in different tumors, observed both experimentally (W-256, PC-1, SM-1) (20, 23) and clinically (breast, urinary bladder and prostate cancers) (24-29), suggest some biological regularity in the impact of the drug on the formation of the pool of these compounds due to hydrolysis of the tumor's structural and functional proteins, as well as free amino acid synthesis and utilization (Fig. 4).

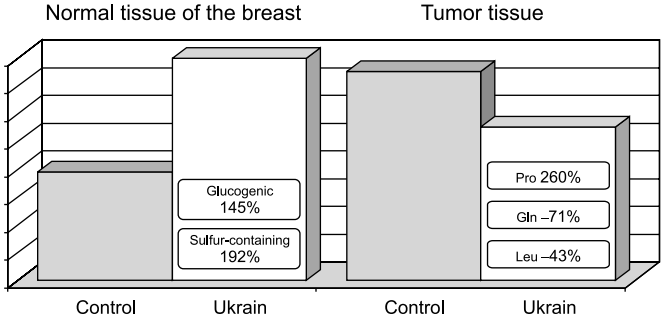

Fig. 4 Influence of Ukrain on amino acid pool in tumor tissue of breast cancer patients.

Particularly noteworthy is the regularly observed elevation of proline concentration by Ukrain, which was significant in all our experimentally and clinically studied tumors. The comparison of the increased amino acid level with the morphological data allowed us to conclude that the activation of proline synthesis or release as a result of Ukrain-induced hydrolysis of proline-containing polypeptides was a biochemical mechanism contributing to the formation of a connective tissue capsule around the tumor, not only facilitating its surgical removal but also preventing growth and metastatic spread.

As has been confirmed by immunofluorescence, Ukrain initially interacts with membrane structures in malignant cells and then is transferred transmembraneously to the nucleus and cytoplasm. This is followed by changes in the levels of tumor amino acids (serine) and related compounds (ethanolamine, phosphoethanolamine), whose conversions are closely linked to the synthesis or degradation of membrane phospholipids (19, 20, 23).

In addition, the interaction of Ukrain with cytoplasmic structures and proteins in malignant cells can simultaneously induce lysosome proteolysis activation and inhibit *de novo* protein synthesis in tumors studied experimentally (30-32).

Our experimental data obtained in studying the activities of the limiting glycolytic, gluconeogenetic and tricarboxylic cycle reactions, as well as the changes in concentrations of the substrates of the main metabolic flows, also suggested that Ukrain interacted with mitochondrial membranes. Therefore, the alterations in the transport and intermediate metabolism of amino acids may be parts of the mechanism of Ukrain's cancerostatic effect.

In addition, the combined decrease in the levels of glutamine and leucine, which are known to be effectors of cell proliferation and protein synthesis, together with increased free amino acid concentrations in the adjacent healthy tissue found after Ukrain administration to patients with cancer of the urinary bladder, may be specific signs of an antitumoral effect and suggest its cancerostatic action is realized *via* regulation of the processes of amino acid pool formation (20).

Analysis of our results according to the principal metabolic criteria establishes the unidirectional effects of the drug under both experimental and clinical conditions.

For example, after the administration of Ukrain in a malignant process, metabolic reaction rates and compound concentrations studied were nearly always restored to normal values, and the resulting picture in tumor-bearer metabolism can on the whole be assessed as one of conventional metabolic comfort.

Our suggestion agrees well with the results of our morphological examination of breast and urinary bladder tumors and the histochemical determination of activities limiting energy production by oxydoreductive reactions in the tumors. The above data indicate that drug administration depleted high-energy compound and nicotinamide coenzyme pools and activated lysosomal enzymes, as well as decreasing the content and disturbing the structure of mitochondria. These processes were also accompanied by DNA fragmentation and angiogenesis inhibition in the tumors (19, 20).

The drug-induced metabolic situation implies activation of gluconeogenetic pathway and redox reactions, as well as inhibition of glycolysis and transamination activities, and energy-producing reactions in the tumor, leading to inhibition of its growth (Fig. 3).

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