THE SURPRISING PROPERTIES OF URIDINE

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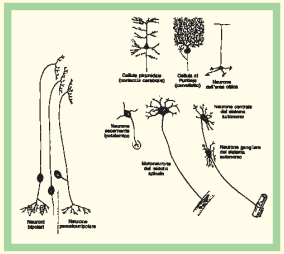
1. Scientific Background.

Uridine is a pyrimidine nucleoside playing a crucial role in mammalian cell life because it participates, together with its phosphate analogs (UMP, UDP and UTP), to protein biosynthesis, cell growth and replication, glucose deposition in stores, disposal of xenobiotics through the glucurono-conjugation pathway, and biosynthesis of cell membranes. Because of this, human cells need a continuous supply of Uridine in order to maintain a healthy life. The diet alone, however, is not able to replenish cells with the needed amount of Uridine, and therefore most cells produce Uridine by themselves, utilizing a biosynthetic process, which includes a bottleneck step inside mitochondria.
Two rare genetic diseases have been described in literature, respectively due to orotic acid accumulation (a precursor of Uridine) or excessive cytosolic 5'-nucleotidase activity. Both diseases lead to decreased availability of intracellular Uridine, with consequent mental retardation or megaloblastic anemia in children, and have been successfully treated with oral Uridine supplementation. Moreover, it has been shown that D-galactosamine and other UTP-depleting substances, which induce acute liver injury followed by cell necrosis due to Uridine shortage, can be counteracted by Uridine administration. Recently, also Mitochondrial disease, a genetic disorder characterized by seizures, muscle weakness and development delays, has been successfully treated with Uridine.
Blood levels of Uridine in humans are tightly regulated, especially by enzymes present in the liver, and are generally found between 3 and 8 micromolar. The circulating nucleoside is actively taken up by cells, easily diffusing into any organ including the brain, and accumulates in cell organelles, normally in the form of pyrimidine nucleotides ready for utilization by specific enzymes. When the external Uridine supply by blood (the so-called "**salvaged**" Uridine) is too low, cells utilize glutamine and carbon dioxide, together with some well-studied enzyme complexes, in order to make Uridine (the "**De novo**" Uridine). This latter biochemical pathway is emerging as a focus for promising pharmacological discoveries, because it has been demonstrated that it is activated by mitogens, and depressed by some well-known anti-tumor, anti-viral and immunosuppressant drugs.
Nobody knows the rate of utilization of the "de novo" or "salvage" Uridine pathway by resting mammalian cells, and it is unclear if there are separate pyrimidine pools inside the cells, preferentially utilizing one or the other path. It has been proposed, however, that the "de novo" synthesis is mostly utilized by rapidly-dividing cells, while the "salvaged" Uridine is especially relevant for the physiological activities of quiescent cells. Strangely enough, no report has been published on the possible correlation of Uridine blood levels with human diseases, even though a drop in circulating Uridine concentration must be suspected every time there are liver disorders.
While the "de novo" Uridine synthesis is pharmacologically impaired by many drugs aimed at reducing cell replication, several human illnesses, and especially the degenerating diseases of the aged people, are possibly due to shortage of the Uridine available to cells. In fact, decreased concentrations of circulating Uridine (due to ageing or to liver problems) are expected to stimulate the "de novo" pathway inside the cells; but this path includes a crucial step inside mitochondria, and it is probably defective in pathological ageing, as it is in genetic mitochondrial diseases. Interestingly, peripheral neuropathies, a common complications not only in diabetes, but also after chronic use of drugs inhibiting the "De novo" Uridine pathway, can be reverted by Uridine supplementation.
Finally, Uridine and its phosphorilated nucleotides have been recently proposed as neurotransmitters, because specific receptors (e.g. $P2Y_2$, $P2Y_4$ and $P2Y_6$), activated by Uridine derivatives, have been found in neuronal and other cells. Results obtained so far suggest that Uridine nucleotides cause depolarization of specific neurons, stimulating release of a number of substances, including catecholamines. Uridine as such seems to have a neuromodulatory role, reducing the effects of excitatory aminoacids in the hippocampus.

2. Classical therapeutic perspectives.

Uridine use in human therapy is not new. In fact, Geiger and Yamasaki published a paper in 1956 showing that the mammalian brain relies on a steady supply of circulating pyrimidines (Uridine and Cytidine) for its electrophysiological activity and for maintaining its carbohydrate and phospholipid content. From this basic work, mixtures of Uridine and Cytidine have been introduced on the market in Italy, starting more 30 years ago, for many neurological indications. As it appears now, Uridine is the true pharmacological agent while Cytidine, being transformed slowly to Uridine in blood, acts as a long-lasting Uridine reservoir.
In the field of genetic disorders, since 1960 small groups of children affected by Orotic Aciduria have been successfully treated with several grams of Uridine a day, in order to avoid the dangerous consequences of the disease, while recently a few childrens affected by increased cellular 5'-nucleotidase activity have been treated with 1 g/Kg a day of Uridine, based on the hypothesis that nucleotidase destroys Uridine, therefore rendering necessary an external supply of the nucleoside. A phase II clinical study is in progress, using Uridine in children with mitochondrial disease, on the ground that genetic damages to mitochondria reduce formation of Uridine by the "De novo" pathway, rendering necessary its supplementation in the diet. In preliminary studies, Uridine showed improvements in the major symptoms of the disease, including reduction of the seizures number and recovery of growth, development, coordination and strenght.
A "rescue" therapy, based on large amounts of Uridine, has been introduced in some American and European hospitals, in order to overcome the toxicity problems due to anticancer pyrimidine compounds like 5-Fluorouracil (5-FU). The "rescue" therapy is based on the fact that millimolar concentrations of circulating Uridine can revert most of the 5-FU side-effects, allowing a better schedule in the use of some anticancer drugs.



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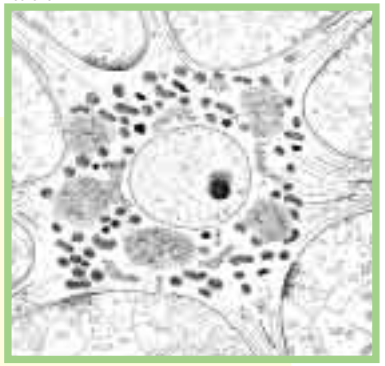
3. New trends in pharmacological uses

A. Uridine in diabetes peripheral neuropathy

Uridine and its nucleotides can have direct actions and modulatory roles on the peripheral nervous system, acting especially on the P2 receptor subtypes like P2Y2, P2Y4 and P2Y6. However, the most important effects of Uridine seem to be related to its metabolic role inside the cells.
Uridine and insulin work together in the building of glycogen stores in hepatocytes, heart and skeletal muscle cells: insulin (like some growth factors) increases the activity of uridine kinase, the enzyme transforming uridine to UTP, which is the rate-limiting component in glycogen biosynthesis and in several metabolic systems leading to the synthesis of essential cell components. Uridine kinase activity is reduced in diabetic rats, and this suggests that UTP shortage might



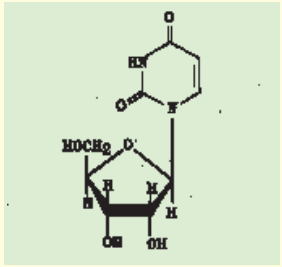
Neurone

be the cause of glycogen depletion observed in the skeletal muscles of diabetic patients. Therefore, uridine administration might revert glycogen depletion, improving the metabolism of peripheral cells.
In rats made diabetic with streptozotocine, Uridine ameliorated nerve conduction with a mechanism believed to be dependent on a trophic effect of Uridine on nerve cells: the spared neurons seem stimulated to grow, in order to replace the died cells, so that the effects on nerve conduction can be maintained also after suspension of the treatment. Previously, in 10 diabetic patients with peripheral neuropathy, Uridine plus Cytidine were given by injection for two weeks, showing increased nerve conduction, while a double-blind, placebo-controlled study on 40 diabetic patients with peripheral neuropathy showed that Uridine alone, by oral route, could improve nerve conduction during the treatment, and also in the follow-up period.

B. Uridine in CNS diseases.

Several studies in the past supported the conclusion that uridine behaves as an anticonvulsant in some seizure models, and might interact with benzodiazepines on GABA receptors, so that uridine itself, or its derivatives, could be developed as anxiolytic, hypnotic or anticonvulsant drugs.
On the other hand, there is a long tradition in Italy on the use of Uridine plus Cytidine, by injection,

in the treatment of important brain damages (traumas and cerebrovascular diseases), in order to reduce injuries and promote functional recovery.



Recent "in vitro" results on cultured neurons suggest that uridine is utilized by cells as a growth factor, inhibiting apoptosis of neurons and promoting differentiation and growth of the survived cells.
Due to the presence of active uridine transport through any kind of cell membranes, and the fact that the compound is easily transported through the blood-brain barrier, Uridine given exogenously by oral route might be proposed for the treatment of important degenerating diseases of the CNS (Alzheimer Disease, Stroke, ALS, and so on).

C.Uridine in the Parkinson's Disease.

Several animal studies suggest that Uridine has a modulatory role on the release of dopamine from dopaminergic brain areas. Chronic uridine administration to rats modulates Dopamine transmission inside the CNS, with a mechanism possibly involving specific effects on Cholecystokinin (CCK) biosynthesis and a facilitated release of cytoplasmic Dopamine pool in the brain. When administered for six months to rats in the drinking water, uridine increased CCK immunoreactivity in nerve terminals of most of the examined telencephalic brain areas; it enhanced turnover rate of spiperone-labelled Dopamine receptors and reduced haloperidol-triggered Dopamine release. Stereotypy and catalepsy, which are side-effects induced in aged rats by dopaminergic and antipsychotic drugs, were inhibited by chronic uridine administration.
Based on the trophic effects showed by uridine in cultered neuronal cells, the drug could constitute a new approach in the treatment of the Parkinson's Disease, because it could improve the symptoms of the disease, due to its dopaminergic effect, and in the same time it could promote regrowth of surviving cells in the substantia nigra, producing therefore long-lasting effects in the outcome of the disease.

D. Uridine in liver disorders

Uridine appears as the key player in the biochemistry of liver, resulting utilized by hepatic cells both in the process of glucose storage (glycogen-synthesis) and in the elimination of most endogenous and exogenous dangerous chemicals (glucurono-conjugation).
The intervention of uridine in the biosynthesis of glycogen suggests intriguing relationships between uridine and insulin in diabetes: Uridine protected the rat brain from hypoglycaemia induced by insulin and, as already said, it reverted the impairment of nerve conduction observed in diabetic patients; therefore, interplays between uridine and insulin are probably not restricted to the liver or to glycogen synthesis.
On the front of xenobiotic disposal, it is known that several endogenous compounds (e.g. bilirubin, steroids, tiroxine) and many drugs, including acetaminophen, valproic acid and benzodiazepines, are metabolized in liver through the glucurono-conjugation pathway, and in experimental animals it has been shown that some chemicals (e.g. galactosamine) cause a liver disease that morphologically resembles the drug-induced hepatitis in humans, which appears related to depletion of uridine in the liver and can be reverted by uridine administration. So, adequate levels of uridine in the liver appear necessary for maintaining the optimal activity of the organ in vital processes.
Furthermore, the liver is the most important reservoir for circulating uridine, which appears stored in hepatocytes after meals, and gradually released into blood for maintaining the physiological cell activities. Any damage to the liver, therefore, will result in impaired uridine distribution throughout the body and eventually in reduced biochemical activities in several organs. Conversely, the rat regenerating liver showed a much increased uridine uptake respect to a resting organ, indicating that uridine storage is a very early and important duty when liver starts healing.

4. Conclusions

The data available in literature support the concept that Uridine fulfils vital roles in liver, as well as in many other organs. A Uridine shortage might be suspected in cases of hepatitis induced by drugs or other toxic compounds, but also in chronic diseases treated with drugs specifically inhibiting the "De novo" biosynthetic route of Uridine (antitumour, antiviral and immunosuppressant drugs), and in many degenerating diseases of the aged people, when mitochondrial activity is impaired. While the diet alone does not seem able to correct the deficiencies of circulating Uridine, its use by oral administration as a supplement to foods could greatly reduce the damages, as demonstrated in children affected by genetic disorders and in tumour patients subjected to the "rescue" therapy with Uridine.