PQQ, a new vitamin as a preventive against heart attack and strokes

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Pyrroloquinoline quinone (PQQ), also known as methoxatin, is a new coenzyme discovered initially in methanobacteria. This small molecule has attracted the attention of many synthetic organic chemists. It is widely distributed in nature and has been found in common fruits, vegetables, milk and egg. Its deficiency in mice causes their slow development and results in fragile blood vessels. It is being considered as a new vitamin of the vitamin-B complex group and may have use in preventing heart attacks and strokes.

Discovery

"New planets are not discovered every day neither are new coenzymes"1. Pyrroloquinoline quinone (PQQ) (I) or methoxatin2 is a new coenzyme found in some bacterial dehydrogenases. Studies have also shown its presence in mammals. There were indications, in the literature, as early as in mid 1960’s, regarding a new cofactor in some bacteria. Only after an ESR study established its o-quinone structure that it was recognized as a new coenzyme3. Based on X-ray diffraction studies4 of this water soluble, deep purple colored coenzyme, was established to be 4,5-dihydro-4,5-dioxo-lH-pyrrolo[2,3-f]quinoline-2,7,9-tricarboxylic acid (I). It is also named as methoxatin since it is involved in the conversion of methanol into formaldehyde catalyzed by methanol dehydrogenase.

Leading synthetic chemists5-9 have approached the synthesis of this coenzyme from different perspectives and this molecule has provided enough challenges. A new and a very short synthesis has been developed by one of us10. The synthesis of PQQ analogues has also attracted attention11,12. "Kilogram Manufacture of PQQ" has also been discussed13. The synthetic material is marketed by Sigma-Aldrich, while PQQ obtained from bacterial sources is marketed by Mitsubishi Gas Chemical Company and by Wako Pure Chemical Industries. At present it is costly and its price will have to come down considerably, if it is to find wide acceptance as a vitamin.

Mechanism of action

Though PQQ is an o-benzoquinone, it functions in biochemical systems as a p-benzoquinone (loc. cit.). Studies have shown that the cofactor is catalytically active only in semiquinone form14. The mode of action of the coenzyme involves reduced form of PQQ (PQQH2), product release, followed by reoxidation to PQQ via a semiquinone radical, and two sequential single electron transfers to cytochrome CL15. There are reports that calcium (Ca2+) plays an important role in its activity16.

Distribution in nature

It is widely distributed in oxidases and dehydrogenases from a variety of bacteria17,18, plants19, and mammals20-22. PQQ is found in most common foods; especially tea, papayas, and kiwi fruits, in large amounts23. There are evidences that it is also found in mammalian enzymes. Recent evidence suggests that PQQ is an essen-
tial nutrient for mouse pups\textsuperscript{24}. The PQQ-deprived mice showed slow development, friable skin, fragile blood vessels, reduced immune response, and poor reproductive performance (loc. cit.). PQQ seems to be an important nutrient for rodents\textsuperscript{25a}. Furthermore, PQQ, given to animals in pharmacological amounts, affords protection against hepatotoxicity caused by liver poisons in rats; acetaldehyde accumulation following ethanol loading in rats; oxidative-stress-induced cataract formation in hydrocortisone-treated chick embryos; neuroexcitatory agents like N-methylaspartic acid that target the glutamate receptor redox site in neurons\textsuperscript{26}.

At the molecular level, PQQ is involved in the degradation of an essential amino acid lysine\textsuperscript{25b} (Fig. 1). The symptoms observed in PQQ-deprived mice are likely to come from the inhibition of lysine degradation or as yet undiscovered PQQ-dependent pathways. Recent evidence indicates that PQQ, first found as a bacterial cofactor for alcohol dehydrogenase, is widely distributed in animal cells, tissues, and fluids and also functions as redox cofactor in mitochondrial complex (loc. cit.).

\begin{table}
\centering
\begin{tabular}{|c|c|c|}
\hline
Foods & ng/g or ng/ml & Foods & ng/g or ng/ml \\
\hline
Broad bean & 18 & Orange & 7 \\
Soyabean & 9 & Papaya & 27 \\
Potato & 17 & Green tea & 30 \\
Parsley & 34 & Oolong tea & 28 \\
Cabbage & 16 & Whiskey & 8 \\
Carrot & 17 & Wine & 6 \\
Celery & 6 & Sake & 4 \\
Green pepper & 28 & Natto & 61 \\
Spinach & 22 & Tofu & 24 \\
Tomato & 9 & Miso & 17 \\
Apple & 6 & Milk & 3 \\
Banana & 13 & Egg (yolk) & 7 \\
Kiwi fruit & 27 & Egg (white) & 4 \\
\hline
\end{tabular}
\caption{Distribution of PQQ in some common food items}
\end{table}

The distribution of PQQ in common fruits, vegetables, cereals, milk and egg has been described in Table 1\textsuperscript{27}.

Granting vitamin status

In the last two decades, there has been an increasing acceptance of PQQ as a vitamin. The process of a compound being given a status of vitamin varies from country to country. Questions are already being raised whether it should be included in the vitamin B complex tablets along with niacin/nicotinic acid and riboflavin (vitamin B2), being sold in the market. Recent studies have proposed that PQQ should be granted the status of a new vitamin (loc. cit.) and should be included in vitamin B group (loc. cit.).

PQQ and electrochemistry

PQQ dependent enzymes catalyze the oxidation of a wide range of primary alcohol due to the unique electrochemical characteristics of this coenzyme, which are described in several papers. These include potentiometric titration and cyclic voltammetric studies\textsuperscript{28}. Based on measurements using cyclic voltammetry at a cystamine modified gold (Au) electrode, the redox potential of PQQ (\(E^0\)), has been found to be \(-0.125\) V vs SCE (at pH \(= 7.0\)) (loc. cit.). A PQQ monolayer has been assembled on an (Au)-electrode surface by carbodiimide coupling of the PQQ carboxylic groups to the amino groups of the cystamine monolayer (The reversible electrochemical process has been found for the PQQ-monolayer) (Fig. 3). Since the immobilized PQQ molecules are fixed at the electrode surface via the cystamine monolayer, the electrochemical process remains reversible even at pH \(> 7\).

Based on the observation that the electro catalytic oxidation of thiols at a glass carbon (GC) electrode coated with a single-wall carbon nanotube (SWNT) film, a PQQ/SWNT/GC electrode has been successfully applied for the assay of both l-cysteine and N-acetyl-l-cysteine in the dietary supplements\textsuperscript{29}. Studies based on the electrochemistry of PQQ and its coupling with glucose oxidase at different pH values has as led to the construction of PQQ-glucose oxidase-modified enzyme electrodes for the de-
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Fig. 2. Stepwise assembling of a PQQ-modified gold (Au)-electrode (taken from Ref. 27).

Fig. 3. Cyclic voltammograms of a PQQ-modified Au-electrode at different potential scan rates: (a) 100 mV s\(^{-1}\), (b) 50 mV s\(^{-1}\), (c) 25 mV s\(^{-1}\) (taken from Ref. 27).

tection of glucose\(^{30}\). PQQ modified electrodes can be used to prepare biosensors based on PQQ dependent enzymes to facilitate chemical reactions typical of PQQ directly on the electrode surface, as these electrodes exhibit very high stability (loc. cit.). A biofuel cell, which has been shown to operate at an open circuit potential of 1.0 V, has a maximum current of 7.5 mA, and a maximum power of 4.07 mW/cm\(^2\), has been developed employing PQQ-dependent alcoholic dehydrogenase immobilized at the bio-anode. Tests showed that this biofuel cell has an overall lifetime upwards of 200 days\(^{31}\). A review on the redox reactions of pyrroloquinoline quinone (PQQ), oxidative decarboxylation of amino acids by PQQ, and the use of PQQ enzymes as biosensors has also been published\(^{32}\).

Future trends

We are interested in the synthesis of PQQ and its analogues as coenzymes for enzymes which mediate the continuous reduction of oxidized hemoglobin in erythrocytes and further protect against re-oxygenation injury in both the heart and the brain. "PQQ also diminishes brain necrosis in a rat model of a stroke"\(^{33}\). Thus PQQ and PQQ analogues may possess the potential for use as therapeutic agents in preventing heart attacks and brain stroke.

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