KOJIC ACID AND ITS DERIVATIVES: HISTORY AND PRESENT STATE OF ART

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SUMMARY

Kojic acid (5-hydroxy-2-hydroxymethyl-4-pyranone) represents an attractive polyfunctional skeleton for development of biologically active compounds. The authors prepared a great variety of kojic acid derivatives and selected biological properties have been studied. Thus, kojic acid derivatives are promising compounds that might advantageously be used in human and/or veterinary medicine and also in preparation of new, even more biologically active preparations.

Key words: kojic acid, 5-hydroxy-2-hydroxymethyl-4-pyranone, derivatives, chelating agent, fungicide, inhibitors of neoplastic cell growth

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INTRODUCTION

Kojic acid was discovered in Japan by Saito in 1907, who isolated that "new" compound from mycelium of the fungus *Aspergillus oryzae* grown on steamed rice ("koji" in Japanese), and this name was given to that organic compound by Yabuta in 1913. Later in 1924, Yabuta succeeded in clarifying the structure of kojic acid, and introduced that fungal metabolite as 5-hydroxy-2-hydroxymethyl-4-pyranone (1). Kojic acid can be produced from various carbohydrate sources in an aerobic condition by a variety of microorganisms. Its lethal dose (LD_{50}) evaluated in mammals was found to be approximately 1 g/kg (2). Kojic acid contains a polyfunctional heterocyclic, an oxygen containing skeleton with several important reaction centres enabling additional reactions, oxidation and reduction, alkylation and acylation, substitution nucleophilic reactions, a ring opening of the molecule, substitution electrophilic reactions, and finally chelation.

A weak bacteriostatic effect of kojic acid reported by Jennings and Williams (1) causes that many research laboratories searching for new antibiotics focused their attention on other, more perspective fungal metabolites. Some of kojic acid physicochemical and biological properties were "rediscovered" in the laboratories of the Slovak Technical University and the Slovak Academy of Sciences in Bratislava, and twenty years later, we succeeded in preparation of novel two mutant strains of fungus *Aspergillus tamarii* (CCM-F-780, CCM-F-781) producing kojic acid in larger quantities when compared to different species of wild strains of *Aspergillus tamarii* (3, 4).

Protocols for the fermentation process utilizing various carbohydrates containing media and the CCM-F-780 or CCM-F-781 kojic acid producing mutant strains have been established (5), and also novel data on enlargement of the kojic acid production by fermentation process were reported (6). The fermentation process comprises several well-known steps starting from inoculation cultivation media by kojic acid producing microorganisms resulting in yielding pale white-yellow prismatic needles of kojic acid.

Since it is freely soluble in water, ethanol, acetone or ethyl acetate, kojic acid was advantageously employed for the preparation over one hundred and fifty various kojic acid derivatives, a part of them even represented new chemical individuals, never synthesised before (7, 8). Physicochemical properties (hydrophobicity, acidity, metal complexing ability), which are expected to play a significant role in biological activity of kojic acid derivatives were determined for a reasonable number of compounds by quantitative structure-time-activity relationship methods (QSTAR) with the aim to predict properties of new derivatives and also to obtain the data for investigation of the quantitative structure-time-activity relationships (9–11).

ANTIFUNGAL ACTIVITY OF 4-PYRANONE HALOGENDERIVATIVES

We have reported a new class of fungicides that may be used as principal constituents of biologically active preparations and have brought evidence showing significant antifungal effects of 5-hydroxy-2-chloromethyl-4-pyranone, 5-hydroxy-2-bromomethyl-4-pyranone and 5-hydroxy-2-iodomethyl-4-pyranone on variety strains of dermatophytes and/or micromycetes (12, 13). Kojic acid does not exert any antifungal activity, however, copper (II) salts of kojic acid halogenderivatives were found to be even more active than halogen-substituted kojic acid derivatives, and

thus they might serve as principal constituents for therapy in human or veterinary dermatology. Novel azidometalkojates (Cu, Mn Mg, Zn and Ni salts of 5-hydroxy-2-azidomethyl-4H-pyran-4-one) as well as derivatives of 4H-pyran-4-one derivative with the atom of sulphur in the side chain of the molecule have been prepared at the Slovak Technical University. It has been shown that the nickel derivative of the azidokojate exerts the strongest antifungal activity when compared to other newly prepared azidometalkojates (8). Similarly, several sulphur-containing derivatives of kojic acid were found to exert antifungal activity 16).

ANTIINFLAMMATORY AND ANTINEOPLASTIC EFFECTS OF KOJIC ACID DERIVATIVES

It has been shown that kojic acid may exert slight anti-inflammatory effects (17) that may advantageously enhanced by subsequent derivation of selected kojic acid derivatives (18). In search for new compounds possessing antitumour activity, we examined the effects of kojic acid halogen derivatives on the proliferation of both the leukemia L 1210 cells and the rat pituitary GH_4C_1 tumour cells. We have reported for the first time that a group of several halogen derivatives of 5-hydroxy-2-hydroxymethyl-4-pyran-4-one may act as promising drugs with antileukemic activity (19). Moreover, we have found that the effect of 5-hydroxy-2-halogenmethyl-4-pyran-4-one derivatives was not due to the metal ion chelating ability. Two halogen derivatives of kojic acid (5-hydroxy-2-chloromethyl-4-pyran-4-one and 5-hydroxy-2-bromomethyl-4-pyran-4-one) were found to inhibit DNA, RNA and protein synthesis (20). 5-benzyloxy-2-thiocyanatomethyl-4-pyran-4-one at 2.6 μM was found to inhibit significantly neoplastic cell growth as well as it inhibits DNA synthesis and cytoplasmic phosporylation (21). Also, the antineoplastic/cytotoxic effect of selected azidometalkojates (Cu, Zn, Mn, Mg and Ni salts) was evaluated on HeLa cells, and the highest antitumour effect was demonstrated by zinc salt of the azidokojates (8). On the other hand, we have also tested the polarographic behaviour and potential carcinogenity of kojic acid derivatives. The most of kojic acid derivatives prepared by our research group was found to possess a very low or almost no potential carcinogenity (22). Further novel compounds, selenocyanato-kojic acid derivatives have been synthesized in our laboratory and their effects on neoplastic cell growth studied (23). Since, the above mentioned novel selenium containing kojic acid derivatives and their effects on neoplastic cell growth in vitro have been found very recently, the further investigation on its, especially, in vivo biological activity, is highly desirable.

OTHER BIOLOGICAL EFFECTS OF KOJIC ACID OR ITS DERIVATIVES

Kojic acid is known to inhibit the catecholase activity of tyrosinase, which is the rate-limiting essential enzyme in the biosynthesis of the skin pigment melanin. Melanocyte treated with kojic acid become nondendritic with a decreased melanin content due to reaction of kojic acid with metals (24). Thus, kojic acid is considered to be a powerful skin lightening/depigmenting agent using externally in skin care products. It has also been shown that kojic acid enhance significantly neutrophil phagocytosis and lymphocyte proliferation by phytohemagglutinin, it enhances a number of leukocytes activities while scavenging reactive oxygen species generated in tissues or blood (25). More than twenty years ago, we demonstrated that 5-acetoxy-2-acetoxymethyl-4-pyran-4-one was able to inhibit *Drosophila melanogaster* development. On the other hand, no insecticidal activity of kojic acid was observed (26, 27).

Kojic acid is a weak mutagen in bacteria, and was found to be nonmutagenic in eukaryotic system either in vivo or in vitro (28). Human intoxication from consumption of oriental fermented foods containing kojic acid has not been reported, however, there are still inconsistent and controversial results on kojic acid toxicity (29). Kojic acid at high doses is toxic. In F344 rats or (C57BL/6N x C3H/N)F1 mice, high doses of kojic acid in food interrupts thyroid function, primarily by inhibiting iodine uptake. It causes a decrease of serum 3,5,3´- triiodo-L-thyronine and L-thyroxine, and subsequently, leads to development of diffuse hyperplasia and follicular adenomas (30, 31).

In conclusion, we are optimistic that future studies on biological properties of kojic acid derivatives may lead to the development of a new class of specific and effective pharmaceutical agents.

Acknowledgement

This work was supported in part by the grant of VEGA No. 2/2070/22.

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