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Original Article

A CRITICAL INSIGHT INTO SHIKIMATE KINASE PATHWAY

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ABSTRACT

Objective: Tuberculosis is the most infectious disease that appears to be dreadful even in the presence of anti tubercular drugs. The problem of MDR-TB is growing at an alarming rate and the prevalence of the disease cause devastation to the molecular level. We hereby carry out a review of shikimate pathway used for tuberculosis management. Based on the available evidence on its vital roles, we highlight ways in which their therapeutic potential can be properly harnessed for possible integration into the country's healthcare system.

Methods: Information was obtained from a literature search of electronic databases such as Google Scholar, Pubmed and Scopus up to 20115 for publications on shikimate pathways and their therapeutic targets for Multi Drug Resistance Tuberculosis (MDR-TB).

Results: Numerous factors have been reported to be the causative agent in the progression of the disease. Apart from this, a number of transcription factors is also been involved in the down regulation of the modulatory pathology. The emergence of MDR-TB washes out the treatment and control of Tuberculosis at an extremely difficult stage. Epidemilogical data revels that Tuberculosis kill approx. 3 million people in a year. Shikimate Kinase and other agents can be involved therapeutic target and evaluation of new pathways. Emphasis need to be urgently given for the diagnosis and treatment of the TB in the society effectively.

Conclusion: This review, therefore, provides a useful resource to enable a thorough assessment of the profile of Shikimate Kinase Pathway used in MDR-TB management so as to ensure a more rational use. Shikimate Kinase is one of the main enzymes involved in Shikimate pathway that has emerged as a vital target in many of the morbidity.

Keywords: Tuberculosis, MDR-TB, Shikimate Kinase, Shikimate pathway.

INTRODUCTION

Multidrug-resistant tuberculosis (MDR-TB) is an emerging problem in many parts of the world, and levels of MDR-TB among new TB patients are increasing in sub-Saharan Africa. HIV has not been reported as a risk factor and there are no reports of the statistical association between spoligo type and drug resistance pattern. Increased capacity for diagnosis and treatment of MDR-TB is needed, with an emphasis on recurrent TB cases and refugees [1].

Although second-line anti-tuberculosis (TB) injectable drugs have been widely used to improve treatment outcomes of multidrug-resistant TB (MDR-TB), little is known about the prevalence and mechanism of second-line injectable drug resistance among MDR *Mycobacterium tuberculosis* isolates in China. Individualized drug-susceptibility to three major second-line injectable drugs is essential in order to generate more effective treatment regimens for MDR patients [17]. The emergence of multidrug-resistant tuberculosis (MDR-TB) and extensively drug-resistant tuberculosis (XDR-TB) makes the treatment and control of tuberculosis difficult. Rapid detection of drug-resistant strains is important for the successful treatment of drug-resistant tuberculosis; however, not all resistance mechanisms to the injectable second-line drugs such as amikacin (AK), kanamycin (KM), and capreomycin (CAP) are well understood [15].

The aromatic compounds such as aromatic amino acids, vitamin K and ubiquinone are important prerequisites for the metabolism of an organism. All organisms can synthesize these aromatic metabolites through shikimate pathway, except for mammals which are dependent on their diet for these compounds.

Aromatic amino acids, vitamin K and ubi quionones serve as an intricate role in the metabolic pathways of organisms. The synthesis of the aromatic metabolites is largely dependent on shikimate pathways, thus this serve as a mandate part in the various metabolic functioning.

The amino acid sequence and x ray crystallography study help the researchers to develop a wide variety of compounds that have shown protective role in combating against various pathogens [11].

The aromatic amino acids like tryptophan, phenylalanine and tyrosine apart from playing their intricate role in the synthesis of proteins, also play a dictating role in the synthesis of secondary metabolites that serve an indispensable role in human nutrition and health. Thus, the role of shikimate kinase pathway cannot be ignored as numerous therapeutic pathways have a modulatory part associated with this pathway [12].

Tuberculosis (TB) remains the leading cause of mortality due to a bacterial pathogen, *Mycobacterium tuberculosis*. However, no new classes of drugs for TB have been developed in the past 30 y. Therefore there is an urgent need to develop faster acting and effective new antitubercular agents, preferably belonging to new structural classes, to better combat TB, including MDR-TB, to shorten the duration of current treatment to improve patient compliance, and to provide effective treatment of latent tuberculosis infection. The enzymes in the shikimate pathway are potential targets for development of a new generation of antitubercular drugs. The shikimate pathway has been shown by disruption of aroK gene to be essential for the *Mycobacterium tuberculosis*. The shikimate kinase (SK) catalyses the phosphorylation of the 3-hydroxyl group of shikimic acid (shikimate) using ATP as a co-substrate. SK belongs to family of nucleoside monophosphate (NMP) kinases. The enzyme is an alpha/beta protein consisting of a central sheet of five parallel beta-strands flanked by alpha-helices [25].

The metabolism of carbohydrates leading to production of aromatic compounds is linked by shikimate pathway. The intermediate produced by these pathways serve as important substrates and play a modulatory role on the other pathways also. The shikimate pathway is found only in microorganisms and plants, never in animals. All enzymes of this pathway have been obtained in pure

form from prokaryotic and eukaryotic sources and their respective DNAs have been characterized from several organisms [13]. Lignin is a major component of plant secondary cell walls.

The enzymes of the shikimate pathway represent potential molecular targets for the development of non-toxic antimicrobial agents and anti-parasite drugs. One of the most promising of these enzymes is shikimate kinase (EC 2.7.1.71), which is responsible for the fifth step in the shikimate pathway [18].

Tuberculosis remains one of the most dreaded infectious diseases notwithstanding the availability of a number of anti-tuberculosis drugs. The recent rise of multidrug-resistant tuberculosis and its association with HIV infection poses a challenging health concern. Therefore, there exists a pressing requirement to identify novel drug targets and develop new anti-tuberculosis drugs that will be effective against multidrug-resistant-tuberculosis. Shikimate kinase is a novel and attractive drug target as it is vital for the survival of *Mycobacterium tuberculosis* but is absent in mammals. Hence, inhibitors designed against shikimate kinase will be specific to the pathogen and be least harmful to the host [20].

Worldwide, tuberculosis (TB) remains the most frequent and important infectious disease causing morbidity and death. One-third of the world's population is infected with *Mycobacterium tuberculosis* (MTB), the etiologic agent of TB. The World Health Organization (WHO) estimates that about eight to ten million new TB cases occur annually worldwide and the incidence of TB is currently increasing. In this context, TB is in the top three, with malaria and HIV being the leading causes of death from a single infectious agent, and approximately two million deaths are attributable to TB annually. In particular, pulmonary TB, the most common form of TB, is a highly contagious and life-threatening infection. Development of drugs which display lasting antimycobacterial activity *in vivo* is desirable, since they can be administered with long intervals and consequently facilitate directly observed therapy and enhance patient compliance. Development of novel antituberculosis compounds to combat MDR-TB is urgently needed. The eradication of slowly metabolizing and, if possible, dormant populations of MTB organisms that cause relapse, using new classes of anti-TB drugs is very promising for prevention of TB incidence, because it will markedly reduce the incidence of active TB from persons who are latently infected with MTB.

Immuno adjunctive agents, especially ATP and its analogues, which potentiate macrophage antimycobacterial activity via purinergic P2 receptors. The aim of this symposium is to address the future prospects of the development of new drugs and drug regimens for anti-TB chemotherapy. There are a number of difficulties in drug-design for the development of new drug formulations with increased potential for antimycobacterial effects, excellent pharmacokinetics, and tolerability. It should be emphasized that the most urgent goal of chemotherapy of TB and MAC infections, especially which associated with HIV infection, is to develop highly active, low-cost drugs which can be used not only in industrialized countries but also in developing countries, since the incidences of AIDS-associated intractable TB and MAC infections are rapidly increasing in the latter. The development of new antitubercular drugs is crucial. Developing inhibitors of shikimate kinase (SK) in the shikimate pathway will provide a selective target for antitubercular agents [22].

Modulators of shikimate kinase pathway

Shikimic acid was first isolated from *Illicium religiosum* in 1885 in Japan and was named after plant, shikimi-no-ki. After the discovery, it has shown to play an important part in the synthesis of many important aromatic compounds. It helps in the synthesis of compounds like folic acid and other compounds exhibiting aromaticity. Thus it has emerged as an intricate plant in the various metabolic pathways. Only a limited number of plant phenols have aromatic rings that are synthesized through another mechanism, that is, the polyketide condensation of acetate units.

Shikimic acid has been found to occur in many tissues of a variety of plants, with a sufficiently high percentage. The accumulation of shikimic acid depends on the rate of synthesis in plants. Thus it is highly variable and gives birth to the wide variety of compounds. In addition, shikimic acid is observed to accumulate in those tissues wherein the metabolic processes are stopped or slow, such as storage tissue of seeds and fruits [23].

Plants as a raw material for shikimate kinase

The first experiments on isolation and recovery of high-purity shikimic acid were conducted in the 1960s, twentieth century. For example, Weinstein *et al.* have devised an effective technique for isolating C14-labelled shikimic acid from *G. biloba* L. for the purpose of studying the metabolism in plants, particularly for studying the shikimate pathway. The plant was kept for several days for metabolism in an atmosphere of radioactive carbon dioxide. The plant material was extracted with ethanol and water, and the extract was passed through a Dowex 50-X4 (H^+form) column to remove main impurities. The eluate was then passed through a column of Dowex 1-X8 (acetate form). The gradual elution with an aqueous acetic acid provided an excellent separation of shikimic and quinic acids. The final passage through the Dowex 1-X8 column gave compounds sufficiently pure for direct usage. Two hundred eighty-seven grams of fresh leaves gave 2.12 g of shikimic acid (0.74% on a wet basis) and 0.593 g of quinic acid.

The shikimic acid content in G. biloba L. is known to be no less than 4% based on wet leaves. Accordingly, the aforesaid technique gave an extremely low yield (below 20%), which is due to the high shikimic acid loss during chromatographic purification.

Thus, there are four general approaches of producing shikimic acid: (1) synthetic approach oriented on the chemical synthesis of shikimic acid (omitted in the present review); (2) an approach where plants serve as shikimic acid sources; (3) an approach where microorganisms are shikimic acid sources; (4) enzymatic approach where shikimic acid synthesis involves enzymes.

The enzymatic technique of producing shikimic acid from quinic acid can be attractive only as a laboratory-scale synthesis. This method requires a cheap source of quinic acid; it is multistage and needs the cultivation of microorganisms and isolation of enzymes there from. It is therefore very labor-intensive, which will cause a rise in price for the product when produced on a commercial scale. Hence, the enzymatic method cannot be applicable to the industrial production of shikimic acid at the present stage of development of biotechnology in the world [23].

All the techniques to produce shikimic acid by its isolation from plants have a number of limitations for use in industry. Their main drawback is that they are confined to the region where the plants grow. Moreover, the isolation of shikimic acid from plants is limited by a certain season of year when the shikimic acid level is sufficient for industrial process. In contrast to methods that utilize plant parts as raw materials, the culture broth-based techniques for isolating shikimic acid are free from such limitations. Disadvantages of the microbiological methods include high labor intensiveness bound up with cultivation of microorganisms and complexities in scaling up. Nevertheless, they have an undisputable advantage over the methods utilizing plants as raw materials, that is, high yields of shikimic acid and a relative purity and homogeneity of raw material. Microbes do not require huge areas for seeding, and their rates of growth and metabolism have no parallel in the vegetable world. The isolation of shikimic acid from culture broths is simple and requires no expensive reagents and sorbents [23].

CONCLUSION

The above paper gives an insight of the various approaches in the context of shikimic acid. It can be produced from the variety of agents some of which are very costly and requires proper care. However, the presence of culture broths enables the formation of shikimic acid which is easy to preapere and relatively inexpensive. The shikimic acid has been used in variety of disorders and thus there is need of exploration of some vital therapeutic agents to cause a global change in the modern era.

CONFLICT OF INTERESTS

Declared None.

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