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In vitro Thrombolytic Activity of a Purified Fibrinolytic Enzyme from *Chlorella vulgaris*

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Abstract

A fibrinolytic enzyme from the microalga *Chlorella vulgaris* was cultivated in mixotrophic conditions (corn steep liquor 1%), with maximum cell concentration and productivity of 1637.45 ± 15 mg L^{-1} and 181.93 mg L^{-1} day^{-1}, respectively. The enzyme was purified by a single chromatographic step, using ion exchange chromatography, with peaks correlated to 2 bands with respective molecular weights of 100.3 kDa and 45 kDa, determined by SDS-PAGE. A fibrin zymography was performed and found that only the 45 kDa band has fibrinolytic activity. After purification, fibrinolytic activity was estimated at 1834.6 U mg^{-1} and 226.86 mm^2, by fibrin plate assays. The enzyme activity was enhanced in the presence of Fe^{2+} and inhibited by phenylmethane sulfonyl fluoride (PMSF) and ethylenediamine tetracetic acid (EDTA), which suggest it to be a metal-dependent serine protease. The extract also showed a red blood cell lysis less than 4% and *in vitro* thrombolytic activity of 25.6% in 90 minutes of reaction. These results indicate that the fibrinolytic enzyme from *C. vulgaris* may have potential applications in the prevention and treatment of thrombosis.

Keywords

Fibrinolytic enzyme; Thrombolytic activity; Corn Steep Liquor; *Chlorella vulgaris*, Thrombosis.

1. Introduction

Cardiovascular diseases are caused by disorders of heart and blood vessels, which include coronary heart disease (heart attack), cerebrovascular disease (stroke), raised blood pressure (hypertension), peripheral artery disease, rheumatic heart disease, congenital heart disease and heart failure. The major causes of cardiovascular dysfunctions are the tobacco use, physical inactivity, unhealthy diet and harmful use of alcohol [1].

Fibrin accumulation is the primary cause of cardiovascular diseases and can lead to morbid and lethal disorders such as acute myocardial infarction, ischemic heart disease and high blood pressure. Many thrombolytic agents, such as tissue plasminogen activator (t-PA), urokinase (UK), and streptokinase (SK) [2], are used to prevent cardiovascular disorders, although they are well reported for their several side effects