Metabolic and biochemical alterations in induced-diabetic rats treated with *Annona muricata* and the associated mechanism using NMR-based metabolomics approach

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Abstract

Patients are increasingly turning to herbs for diabetes treatment, resulting in a rise in demand for alternative therapies derived from plants. In underdeveloped nations, specifically, where most people have limited resources and do not have access to contemporary treatment, plants continue to play a crucial role in the treatment of diabetes. *Annona muricata*, often known as "Soursop" in various regions of the world, is a well-known medicinal plant with multiple applications. It is a common plant used in Africa and South America to manage various types of disease. However, there are few studies that investigate the anti-diabetic properties of this plant and the associated therapeutic process. The anti-diabetic action of *A. muricata* in vitro inspires us to evaluate its in vivo antidiabetic activity. Thus, the antihyperglycemic action of *A. Muricata* was tested on diabetic rats at doses of 200 and 300 mg/kg, and proton nuclear magnetic resonance (^{1}H-NMR) was used to analyse the changes in rats' urine metabolites. The ^{1}H-NMR was also used to evaluate the serum and urinary metabolites changes for the in vivo acute toxicological profile of *A. muricata* leaf ethanol extract. A single 2000 mg/kg dose of *A. muricata* leaf ethanol extract was administered to Sprague Dawley rats over an observational period of 14 days. The toxicity evaluation (physical and behavior observation, body weight, renal function test, liver function test and ^{1}H NMR analysis) showed no abnormal toxicity. Histopathological analysis manifested mild changes, i.e., the treated kidney manifested mild hypercellularity of mesangial cells and mild red blood cell congestion. In addition, there was mild hemorrhage into tissue with scattered inflammatory cells and mild dilated central vein with fibrosis in the liver. However, the changes were very mild and not significant which correlate with other analyses conducted in this study (biochemical test and ^{1}H NMR metabolomic analysis). The diabetic state on rats was achieved by injecting alloxan and nicotinamide. After a course of treatment for 30 days with the standardised extracts of *A. muricata*, the results revealed that the hyperglycemic blood level was lowered to a state that was normal. In addition, the extracts showed a significant management effect on the metabolic parameters of diabetic rats. The results of the metabolomics investigation indicated that this extract managed and modulated the disordered metabolites of diabetic

rats by returning some of them to their normal state. In obese diabetic rats, the extract lowered the serum glucose level and improved the lipid profile. The findings of this study could lead to a better understanding of the molecular mechanism of action of this medicinal plant in the treatment of diabetes. This information may be very beneficial in suggesting the usage of this plant in long-term treatment for diabetes, which may be useful for additional pharmacological and preclinical studies.

Keywords: Biochemical test, toxicity, ^{1}H NMR metabolomics, histopathology, *Annona muricata*, Diabetes