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# Pathological Studies on The Protective Effect of Ginger Extract and Ginger Nanoparticle on Acetaminophen Toxicity in Rats

Submitted by

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**بسم الله الرحمن الرحيم**

**"قَالُواْ سُبْحَانَكَ لاَ عِلْمَ لَنَا إِلاَّ مَا عَلَّمْتَنَا إِنَّكَ أَنتَ الْعَلِيمُ الْحَكِيمُ"**

**البقرة - (32)**

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## Abstract

Acetaminophen (APAP) is widely used analgesics all over the world. However, hepatotoxicity and nephrotoxicity are the most remarkable features of acetaminophen overdose. Ginger is a medicinal plant that has immuno-modulatory, anti-tumorigenic, anti-inflammatory, anti-apoptotic, anti-hyperglycemic, anti-lipidemic and anti-oxidant activities. Recently, ginger extract in form of nanoparticles (NPs) have been investigated in liver protection against alcohol-induced liver damage. We conducted *in vivo* and *in vitro* studies to compare between the protective efficacy of ginger extract and ginger nanoparticles (GNPs) against acetaminophen-induced toxicity from histological and biochemical aspects. *In vivo* study, adult male Sprague Dawley rats were received ginger extract and GNPs orally at dose of 120 mg/kg (3times/week) and acetaminophen at dose of 375mg/kg ($1/10LD_{50}$) daily for three months. Serum Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) as liver functionality indicator were determined. In addition, kidneys functions were assessed by evaluating urea and uric acid levels in serum. Moreover, oxidative stress at the cell level was evaluated by determining malondialdehyde content (MDA) and catalase enzyme (CAT) activity. Meanwhile, histopathological changes in liver and kidney tissues were observed. The present study indicates that liver and kidney biochemical markers are improved in rat pretreated with ginger extract and ginger nanoparticles as well as the activities of cell oxidative stress are statistically significant diminished. In

addition, their histological structure of liver and kidney tissues show very little changes. While rats treated with GNPs demonstrate normal biochemical and oxidative stress marker levels and histological structure relative to ginger extract treated rat.

While *in vitro* study, we use ginger extract and ginger nanoparticles at concentration of 60 µg/ml against hepatotoxicity caused by acetaminophen (APAP) at concentration of 0.1 mg/ml using primary isolated rat hepatocytes. Cytotoxicity was determined by assessing cell viability and leakage of cytosolic enzymes, such as (ALT& AST). Oxidative stress was investigated by measuring levels of CAT and MDA. The cytopathological alterations were studied by light microscope. Exposure of isolated rat hepatocytes to APAP caused cytotoxicity and oxidative injury, manifested by loss of cell viability and significant increase in ALT and AST leakages. As well as, APAP caused progressive depletion of CAT content and increase intracellular MDA accumulation, in addition to alteration in cellular morphology. Pretreatment of hepatocytes with either GE or GNPs ameliorated the hepatotoxicity, oxidative stress and enzymatic leakage induced by APAP. However, GNPs were more effective relative to ginger extract pretreated hepatocytes. From both studies we concluded that, ginger nanoparticles have more protective activities relative to ginger extract.

**Key Words**

Acetaminophen - ginger extract – ginger nanoparticles – oxidative stress- histopathology- isolated hepatocytes.

# *Dedication*

*To my dear parents,*

*To my sisters and brothers,*

*To my daughter Malika and my son Ali,*

*To my husband Mohamed and his family*

*Thanks a lot, I appreciate your great efforts*

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# LIST OF ABBREVIATIONS

| | |
|---|---|
| **ALT** | Alanine aminotransferase |
| **APAP** | Acetaminophen (Paracetamol) |
| **AST** | Aspartate aminotransferase |
| **ATN** | Acute tubular necrosis |
| **BUN** | Blood urea nitrogen |
| **CAT** | Catalase |
| **Ccl4** | Carbon tetrachloride |
| **Cd** | Cadmium |
| **Cox-2** | Cyclo-oxygenase |
| **COX-3** | cyclo-oxygenase |
| **CYP 2E1** | Cytochrome 2E1 |
| **DNA** | Deoxyribonucleic acid |
| **DPPH** | 2,2-diphenlyl-1-picrylhydrazyl |
| **GE** | Ginger extract |
| **GNPs** | Ginger nanoparticles |
| **GSH-Px** | Glutathione peroxidase |
| **Gst pi** | Glutathione S-transferase Pi |
| **H&E** | Hematoxylin and eosin |
| **HRS** | Hepatorenal syndrome |
| **IHC** | Immunohistochemistry |
| **IL-1β** | Interleukin-1β |
| **IL-33** | Interleukin-33 |
| **iNOS** | inducible nitric oxide synthase |
| **MDA** | Malondialdehyde |
| **NAC** | N-Acetylcysteine |
| **NAPQI** | N-acetyl-para-benzoquinone imine |
| **NO** | Nitric oxide |
| **Pb** | Lead |
| **RNA** | Ribonucleic acid |
| **ROS** | Reactive oxygen species |
| **RSTI** | Repeated supratherapeutic ingestion |
| **SGOT** | Serum glutamate oxaloacetate transaminase |
| **SGPT** | Serum glutamate pyruvate transaminase |
| **SOD** | Superoxide dismutase |

| | |
|---|---|
| **SPSS** | Statistical package for social science |
| **TBARS** | Thiobarbituric acid reactive species |
| **TNF-α** | Tumor necrosis factor α |
| **TPA** | 12-O- tetradecanoylphorbol-13-acetate |
| **Vd** | volume of distribution |

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