Chapter 4

Determination of Some Antioxidant Activities in Food Supplement Mixture Fruit Containing Noni (Morinda Citrifolia), Vitamin B3, Zinc, Vitamin B1, Coenzyme Q10 and Chromium

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Abstract

Nitric oxide has important functions in cardiovascular, neurological, immunological and many biological systems). Noni (*Morinda citrifolia*) has been found to have antitumor, antiproliferative, proapoptotic, anti-angiogenesis, anti-migratory, anti-inflammatory and immunomodulatory activities. The aim of this study is to determine some antioxidant activities in food supplement mixture fruit containing Noni (*Morinda citrifolia*), Vitamin B3, Zinc, Vitamin B1, CoenzymeQ10 and Chromium. In this study, antioxidant activities were determined by spectrophotometric method. While the MDA level was found to be 0.0012 (mmol/L); the SOD activity was 1377.483 (U/L); reduced glutathione (GSH) level 725.251(mg/dl); catalase (CAT) activity 1275.035 (U/L); glutathione reductase activity (GR) 1439.041 (U/L), and glutathione peroxidase (GPx) activity were found to be 1215.055 (U/L). Some antioxidant activities such as SOD, GR, GST, GPx, GSH and CAT were found to be high in the noni fruit. Malondialdehyde (MDA) level was found to be low. Antioxidant activities were found to be very high in the noni extract. Noni fruit or its extract can be consumed against oxidative stress. We think that this study will contribute to the literature.

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1. Introduction

Free radicals are unpaired electron pairs. The main free radicals are compounds such as hydroxy (OH-), peroxy (ROO.), and superoxide (O_2-) radical. When free radicals are formed, they become stable. Also, these radicals enter the structure of cells and damage them. As a result of this damage, various may diseases occur. Free oxygen radicals have been implicated in the etiopathogenesis of a number of diseases, according to reports. It has been determined that free oxygen radical damage occurs in diseases such as bladder disease, prostate cancer, sepsis, myocardial infarction, stroke, perinatal hypoxic brain injury, glomerulonephritis, uveitis, various cancers and arthritis in laboratory, clinical and experiments. However, antioxidants protect the cell against these radicals (Sosa et al., 2013; Sayır et al., 2019; Günes et al., 2020; Gündüz et al., 2021).

Nitric Oxide is a gas with the chemical formula N-O. It carries datas at the cellular level and can effectively penetrate the cell membrane thanks to its gaseous structure.

Since the past, the importance and benefits of medicinal plants for human health have been investigated. Plants have very important roles in the treatment and prevention of diseases. Herbal medicines have been developed with the extracts of plants used by the public. Also, people have managed to protect themselves from diseases for years (Mill, 1982).

Morinda citrifolia is rich in nutritional value. It has strong antioxidant activity. It strengthens the immune system and balances acid and base in the body. It contains rich minerals and vitamins. It is also present in various alkaloids, polysaccharides, various enzymes and many components (Akihisa et al., 2007; Zhou and Huang, 2022).

This study's objective is to ascertain some antioxidant activities in food supplement mixture fruit containing Noni (*Morinda citrifolia*), Vitamin B3, Zinc, Vitamin B1, CoenzymeQ10 and Chromium.

2. Analysis Method

Noni (Morinda citrifolia), food supplement mixture containing Vitamin B3, Zinc, VitaminB1, CoenzymeQ10 and Chromium, contains malondialdehyde (MDA), which is an oxidative stress parameter, and superoxide dismutase (SOD), reduced glutathione (GSH), catalase (CAT), glutathione peroxidase (Some antioxidant activities such as GSHPx), glutathione reductase (GR) and glutathione S-transferase (GST) were determined spectrophotometrically.

2.1 Measuring the activity of superoxide dismutase (SOD)

Determination of superoxide dismutase (SOD) activity SOD activity was determined according to the method by Popov et al (Popov et al., 2004).

% Inhibition = [(Blank OD – Sample OD) / Blank OD] × 100

2.2 Measuring the level of reduced glutathione (GSH)

GSH was determined according to the method made by Tietz. 800 μl of phosphate buffer was added to 200 μl of serum. Initial absorbance (OD1) at 412 nm was measured. 100 μl of Ellman reagent was added to the same tube and then the second absorbance (OD2) was measured (Tietz, 1969).

Calculation:

The concentration of glutathione was expressed in units of mmol/g of protein.

C / 1000 = (OD2-OD1) / 13600 x E1 x 5/2 x ½

13600: Molar extinction coefficient of yellow color formed during the

interaction of GSH and DTNB.

E1: If a band with a width greater than 6 nm is used, a derivative extrusion

coefficient is used that corrects for both light path and bandwidth differences.

The width of the tape we use is 2 nm.

It was taken as E1=1 in the calculations.

1000: conversion coefficient to mmol.

C: mmol / glutathione (mg/dl)

2.3 Catalase (CAT) activity measurement

The Aeibi method was utilized in this investigation to ascertain CAT activity. First, 1.4 ml of 30 mM hydrogen peroxide (H_2O_2) was placed in the blank tube and 0.1 ml of phosphate buffer was added to it. 1.4 ml of 30 mM hydrogen peroxide (H_2O_2) was placed in the sample tube and 0.1 ml of sample was added on it and the tubes were vortexed. The absorbance values were then measured twice at 240 nm at thirty-second intervals by the spectrophotometric method (Aeibi, 1948).

Activity account:

Activity= (2.3/ΔX) × [(log A1 / log A2)]

ΔX: 30 seconds

2.3: 1mmol optical density of H H_2O_2 in 1cm light path.

2.4 Glutathione reductase (GR) activity measurement

The activity of glutathione reductase was quantified according to the method of Goldberg and Spooner (Jiang and Zhang, 2002).

2.5 Glutathione S-transferase activity measurement

Glutathione-s-transferase activity was performed using the determination method suggested by Mannervik et al. (Mannervik and Guthenberg, 1981).

2.6 Glutathione peroxidase (GPx) activity measurement

Beutler's method of determination was applied to determine glutathione peroxidase activity (Beutler et al., 1983).

2.7 Measuring the level of malondialdehyde (MDA)

Malondialdehyde, Lipid Peroxidation's final product as a result of the reaction of fatty acids with free radicals, was measured with thiabarbituric acid (Jentzsch et al., 1996). 200 ml of serum was placed in the sample tube. 800 ml of phosphate buffer, 25 ml of BHT solution and 500 ml of 30% TCA were added to it. The tubes were mixed by vortexing and kept on ice for 2 hours. It was then centrifuged at 2000 rpm for 15 minutes. 1 ml of the obtained supernatant was taken and transferred to another tube. Then, 75 ml of EDTA and 250 ml of TBA were added to this mixture. The tubes were mixed by vortex again and kept in a hot water bath for 15 minutes. The tubes were then brought to room temperature. Absorbance values were read in the spectrophotometer at 532 nm.

Calculation of malondialdehyde level:

C=F× 6.41× A

C: Concentration

F: Dilution factor

A: Absorbance

3. Results

While the MDA level was found to be 0.0012 (mmol/L), the SOD activity was 1377.483 (U/L); reduced glutathione (GSH) level 725.251(mg/dl); catalase (CAT) activity 1275.035 (U/L); glutathione reductase (GR)

1439.041 activity and glutathione peroxidase (GPx) activity were found to be 1215.055 (U/L).

Table 1. MDA level and SOD, GSH, CAT, GR and GPx activities

Parameters	
GPx(U/L)	1215.055
MDA (mmol/L)	0.0012
GSH (mg/dl)	725.251
CAT (U/L)	1275.035
SOD (U/L)	1377.483
GST (U/L)	1288.015
GR (U/L)	1439.041

4. Discussion

MDA is a marker reflecting oxidative stress. Also , MDA shows lipid peroxidation (Sudha et al., 2001). Antioxidants fight free radicals and also reduce the effects of free radicals. SOD, CAT, GST, GR (glutathione reductase), GPx (glutathione peroxidase) and GSH are the main antioxidants. Antioxidant enzymes are protective type enzymes that increase their activity under oxidative stress conditions. Antioxidants are required as compensatory mechanisms of oxidative stress. SOD is a very powerful antioxidant enzyme (Güneş et al., 2020). CAT is one of the most important antioxidant agents (Gündüz et al., 2021). GSH level protects the cell against reactive oxygen molecules (Gündüz et al., 2021).

Polyphenols have very strong antimicrobial and antioxidant activity. So, these are natural compounds versus synthetic foods.

Nitric Oxide (NO gas) is a substance that provides the circulatory system throughout the body and also supports energy increase. It is also a molecule of life that helps in many other functions of the body. L-Arginine amino acid is a source of NO. The use of NO reduces intimal thickening in atherosclerosis. Thus, in hypertension, L-Arginine also reduces blood pressure as it increases nitric oxide production. As a result, it reduces the proliferation rate in vascular smooth muscle cells (Moncada and Higgs, 1993).

In literature studies, *Morinda citrifolia* (Noni) has found to alleviate DNCB-induced atopic dermatitis in NC/Nga mice by modulating immune

balance and skin barrier function (Kim et al., 2020). In another study, *M. citrifolia* has reported to have antitumor, antiproliferative, proapoptotic, antiangiogenesis, antimigratory, anticancer, anti obesity, anti-inflammatory and immunomodulatory activities. Additionally, thanks to its many properties, it may be a possibly useful medicinal plant for cancer treatment (Chanthira Kumar et al., 2020; Yilmaz et al., 2020).

Some antioxidant activities such as SOD, GST, GR, GPx, GSH and CAT were found to be high in the noni fruit. On the other hand, level of MDA was found to be low. Antioxidant activities were found to be very high in the noni extract. We think that this study will contribute to the literature. More future clinical studies are needed. Thus, the role of *M. citrifolia* in the treatment of cancer and other diseases may be more important. Noni fruit or its extract can be consumed against oxidative stress.

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