Effect of date extracts on the lymphocyte and monocyte cells

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الخلاصة

. در ت اثير مستخلص ات التم ر عل ى الخلاي ا اللمفاوي ة وخلاي ا أحادي ة الن واة البش رية باستخدام تراكيز مختلف ة م ن المستخلص ات ولم دة ثلاث ة أي ام بواس طة طريق ة (MTT) .
وأظهرت الدراسة انه لايوجد تأثير س ام للمستخلص ات م ن أن واع التم ر المختلف ة عل ى الخلايا اللمفاوية وخلايا وحيدة النواة البشرية. ق د أظه رت النت ائج إن التراكي ز المختلف ة من المستخلصات تقاأنها حماية وه ذا ربم ا يع ود إل ى إن المستخلص ات تحت وي عل ى مواد مضادة لأكسدة في الأنواع المختلفة من التمور.

Abstract

The effect of dates extracts were determined in human peripheral lymphocyte and monocyte cells using different concentrations of the extracts for three days- there term assay (MTT) was employed. There was no cytotoxic effect of the date extracts on human lymphocyte and monocyte cells. The results showed that the ideal concentrations of different date extracts have a protective effects of tamer (Barhi, Zahdi and Khistawy) to the lymphocyte and monocyte cells and may be that because these date extracts contain antioxidants in various type of dates.

Introduction

Previous studies have demonstrated that dates have anticancer activity to various kinds of cancer cell line by using tamer AL-Zahdi (1). The aim of this study to test the cytotoxic and the proliferation effect of different kinds of date
(Barhi, Zahdi and Khistawy) on the lymphocyte and monocyte cells from normal adult. Dates high in natural aspirin also high in fiber; have laxative effect dried fruits, including dates, are linked to lower rates of certain cancers, especially pancreatic cancer (2). The phyto chemicals in dates are thought to be responsible for this reduced risk because of their antioxidant and antiproliferative (used to inhibit cell growth) activities (3). In recent years, an increased consumption of antioxidants has been recommended to present or slow the oxidative stress to cells caused by free radicals.

Material and Methods

Date extract preparation:

A quantity of 20gm Tamer of Barhi, Khistawy and Zahdi were extracted in 250 ml D.W using stirrer at 25c^{o} for 24 h D.W was evaporated using a rotary evaporator at 35 – 40 C$^{o.}$ (1).

Blood collection:

Blood was taken from normal adults by puncture; first the area was wiped with alcohol, and left to dry up. A disposable 5 ml syringe was used, sample of blood were transferred into heparin Ned tubes. This sample was used for lymphocyte and monocyte separation.

The isopaque ficol technique originally described by (4) was used for lymphocyte separation.

Cytotoxicity and proliferation Assay:

The micro culture tetraolium dye (MTT) assay was used to determine lympho and monocyte cell. Briefly, the target cell suspension ($2x10^4$ cells) were added to each well of 96 well flat – bottom micro titer plates (corning glass works, corning, NY, U.S.A.).

And each plate was incubated for 24 hours at 37 C^{o} in a humidified 5% CO_2 atmosphere after incubation, 50 ml of date solution or complete medium for control were distributed in the 96 well plates and each plate was incubated for 72 hours. At 37 C^{o}. Following incubation, 20 ml of MTT working solution (5 mg/ml, Sign on. Chemical Co. St. Louis, MSA) was added to each culture well and the cultures were incubated for 4 hours at 37C^{o} in humidified 5% CO_2 atmosphere.

One hundred ML of is propane (sigma chemical CO_2) supplemented with 0.05 NHCL was added to each well.

The absorbance of each well was measured with a micro culture plate reader
(Microreader, Japan. Interment CO-Ltd, Tokyo, Japan) at 520 nm (5).

Results and Dissections

Cell proliferation was induced by PHA for 24h and determined 72h after incubation of cells with the respective additives of different concentrations of date extracts by MTT test.

After the lymphocyte were incubated with concentration of 5,10,25 and 50 µg/ml, of different date extracts for 72h at 37C^{o}, optical cell densities were measured with a micro plate reader. We tested the treatment groups with date extracts in this experiment compared to the control group which we treated with PHA for 96h. The objective of this experiment was to determine if date extracts increases the mitogenic action of PHA.

The proliferation rate of treatment groups were significantly lower ($p<0.001$) than that of the control group (PHA) as shown in tables (1, 2).

Table (1) proliferation effect of date extracts on the lymphocyte cells

Concentration of date extracts µg/ml	% proliferation rate of Barhi	% proliferation rate of Zahdi	% proliferation rate of Khistawy
5	1.3	1.9	1.25
10	13.94	18.9	10.5
25	16.3	13.0	17.0
50	21.6	25.9	26.3
PHA	93%	93%	93%

Table (2) proliferation effect of date extracts on the monocyte cells.

Concentration of date extracts µg/ml	% proliferation rate of Barhi	% proliferation rate of Zahdi	% proliferation rate of Khistawy
5	7.19	2.5	1.3
10	13.1	16.9	1.4
25	16.3	17.6	14.5
50	17.9	18.4	13.6
PHA	90.8	90.8	90.8

Our study has adopted a micro culture assay based on metabolic reduction of MTT to evaluate the cytotoxic effect of date extracts on human peripheral blood lymphocyte and monocyte

cells. Tetrazolium salt is metabolically reduced by viable cells to yield a blue Formosan product measurable in a multiple scanning spectrophotometer. This technique permitted to evaluate dosed pent – effect. Tables (3,4) shows the cytotoxic effect using different concentrations of date extracts (Barhi, Zahdi and Khistawy) on the lymphocyte and monocyte cells which determined by MTT assays.

Tables (3) cytotoxic effect of date extracts on the lymphocyte cells

Concentration of date extracts µg/ml	% cytotoxicity of Barhi	% cytotoxicity of Zahdi	% cytotoxicity of Khistawy
5	8.3	10.9	14.0
10	4	8.9	10.0
25	3.7	4.9	6.8
50	1.7	3.4	3.16

Table (4) cytotoxic effect of date extracts on the monocyte cells

Concentration of date extracts µg/ml	% cytotoxicity of Barhi	% cytotoxicity of Zahdi	% cytotoxicity of Khistawy
5	-1.1	-2.5	1.4
10	-10.1	-11.6	1.1
25	-11.3	-17.5	-14.0
50	-13.8	-17.9	-12.3

These results show there is no cytotoxic effect of date extracts of different types on the lymphocyte and monocyte cells. That means there is a protective effect of tamer (Barhi, Zahdi and Khistawy) to the lymphocyte and monocyte cells and may be that because these date extracts contain antioxidants in various type date (5, 6).

(7) Demonstrate that the date extract contain anti cancer agent. Aglucan of cellular origin has been isolated from dates (*phoenix dactylifera*. L) Structure of the purified glucan was characterized using derivatisation methods including methylation; periodate oxidation, and a cytolysis. Glucose was found to exhibit potent antitumor activity; this activity could be correlated to their (1-3) – B_D- glucan linkages. All forms of sugar (including date)

interfere with the ability of white blood cell to destroy bacteria (8, 9).

Animal studies suggest diets high in sucrose impair some aspects of immune function (10, 11).

The importance of these effects in the prevention of infections in human remains unclear. The results indicate compounds on human lymphocyte cultures, which was an additional activity to their antimutagenic action in vitro, described previously (1) since the compounds of date extracts (Barhi, Khistawy and Zahdi) are natural, relatively non- toxic products, they could be administered to people as regular diet components, both in order to forestall mutations and to modulate the immune system (12, 13).

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