

سُورَةُ الضُّحَىٰ

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

وَالضُّحَىٰ ﴿١﴾ وَاللَّيْلِ إِذَا سَجَىٰ ﴿٢﴾ مَا وَدَّعَكَ رَبُّكَ وَمَا قَلَىٰ ﴿٣﴾
وَلَلْآخِرَةُ خَيْرٌ لَّكَ مِنَ الْأُولَىٰ ﴿٤﴾ وَلَسَوْفَ يُعْطِيكَ رَبُّكَ
فَتَرْضَىٰ ﴿٥﴾ أَلَمْ يَجِدْكَ يَتِيمًا فَآوَىٰ ﴿٦﴾ وَوَجَدَكَ ضَالًّا
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وَأَمَّا السَّائِلَ فَلَا تَنْهَرْ ﴿١٠﴾ وَأَمَّا بِنِعْمَةِ رَبِّكَ فَحَدِّثْ ﴿١١﴾

Dedication

To ... Whom may God bless me because of her

My Mother

To .. The man above all men. Dr. Abbas Ibrahim

My Father

To... my guardian angel...

My Husband

To ...My shelters through rainy days..

My Sisters and Brothers in law

To ...my investment in life...

My Son

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Abstract

The aim of the present study was to isolation, purification and characterization of *Momordica balsamina* seeds lectin (MbSL) and study the possible modulating effects of the purified lectin on four types of commercial cancer cell lines.

A season fresh of *Momordica balsamina* fruit seeds were brought from urban areas of Sudan (Gadriif and north Kurdofofan states). The lectin was isolated from saline extract by affinity chromatography on alpha agarose lactose matrix. The lectin content which was determined by the method of Lowry was about 1200 mg/100 g dry flour.

MbSL agglutinated all types of human red blood cells (RBCs) with preference toward the O blood group. The lectin also agglutinated mouse, donkey and cow red blood cells but showed no effect on goat erythrocytes.

Lactose was the most potent inhibitor of MbSL hemagglutinating activity, minimal inhibitory concentration (MIC) = 25 mM, followed by galactose, MIC=50 mM, and then arabinose, MIC= 100 mM.

The native molecular weight of lectin detected by gel filtration chromatography was 81 kDa and when examined by SDS-PAGE it was found to be composed of a single subunit of molecular mass around 30 kDa.

The search for sequential identities of the purified lectin was carried out by using BLAST (<http://blast.ncbi.nlm.nih.gov/Blast.cgi>), and the N-terminal of the lectin shared major similarities with reported *Momordica charantia* lectin 1(MCL 1) and found to be a ribosome inactivating protein type II (RIP II).

The activity of the lectin remained stable in the pH range 2-12, and it remained stable below 50 °C without losing its hemagglutinating activity. Above 50 °C lectin activity was gradually lost and was totally inactivated at 90 °C.

MbSL activity slightly decreased against denaturation with urea, with significant drop at 3M.

After evaluation of Cytotoxicity of the purified lectin using the MTT Assay in AGS (Human Gastric Adenocarcinoma), MKN45 (Human Gastric Cancer), U87-MG (Human Glioblastoma) and ECV-304 (Human Urinary Bladder Carcinoma) cell lines, the lectin showed no inhibitory effect on the growth of different types of human commercial cancer cell lines. This identified MbSL as a nontoxic ribosome inactivating protein type II.

In conclusion: a lactose-binding lectin from seeds of *Momordica balsamina* medicinal plant shares a high degree of similarity with other Cucurbitaceae family lectins in term of their physicochemical features including sugar specificity, effect of pH and temperature and urea on lectin stability. MbSL was found to be one of nontoxic RIP II and had no effect on the growth of four types of human commercial cancer cell lines chosen to study the modulating effect of the purified lectin on it.

ملخص الدراسة

هدفت هذه الدراسة لعزل و تنقية و توصيف لكيتين بذور نبتة العيبر بالاضافة الى دراسة اثر اللكتين النقي على اربعة انواع من خلايا بشرية تجارية للسرطان.

تم جمع مجموعة من بذور نبات العيبر من المناطق الصحراوية لبعض الولايات سودانية (القضارف و شمال كردفان) ثم تمت تنقية اللكتين بالطرق الكيمائية المناسبة لذلك، كما تم قياس نسبة البروتين بعد التنقية بطريقة لاوري.

اظهرت النتائج قابلية اللكتين النقي للالتصاق بخلايا الدم الحمراء البشرية و بعض خلايا الدم الحمراء الحيوانية، كما اظهرت النتائج قابلية ارتباط اللكتين لمجموعة من السكريات و بالأخص سكر اللاكتوز، كما تم تحديد الوزن الجزيئي للكتين النقي بطريقتين مختلفتين..

اظهرت النتائج ان نشاط اللكتين النقي يتحمل درجات مختلفة من الحرارة و مستويات متعددة من اختلاف الاس الهيدروجيني كما ان نشاط اللكتين النقي يهبط تدريجيا تحت تأثير زيادة تركيز اليوريا.

وعلى الرغم من تحديد اللكتين النقي كأحد البروتينات التي لديها المقدرة علي تعطيل الريبوسوم الا انه لم يؤدي الي تثبيط او تعطيل نمو الخلايا السرطانية المختارة في الدراسة.

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List of Abbreviations

Abbreviation	Full term
AGS	Human Gastric Adenocarcinoma
BLAST	Basic Local Alignment Search Tool
Con A	Concanavalin A
CRA	Carbohydrate-Recognition Domain
DMSO	Dimethyl sulfoxide
ECV-304	Human Urinary Bladder Carcinoma
MB	<i>Momordica</i> Balsamina
MBL	<i>Momordica</i> Balsamina Lectin
MC	<i>Momordica</i> Charantia
MCL	<i>Momordica</i> Charantia Lectin
MIC	Minimum Inhibitory Concentration
MKN45	Human Gastric Cancer
MTT	1-(4,5 dimethylthiazol-yl)-3,5-diphenyl formazan
PNA	Peanut Agglutinin
RBCs	Red Blood Cells
RIPs	Ribosome Inactivating Proteins
RP-FPLC	Reversed-Phase Fast Protein Liquid Chromatography
SDS-PAGE	Sodium dodecyl sulphate- polyacrylamide Gel Electrophoresis
U87-MG	Human Glioblastoma