



STUDIES ON ANTI-HIV ACTIVITY AND CYTOTOXICITY OF LEAF OF *MORINDA CITRIFOLIA* L

PERIYASAMY SELVAM*

Devaki Amma Memorial College of Pharmacy, Chelembra, Pulliparamaba Post,
MALAPURAM – 673634 (Kerala) INDIA

ABSTRACT

Different leaf extracts of *Morinda citrifolia* L have been studied for antiviral activity against replication of HIV-1 (IIIB) and HIV-2(ROD) in MT-4 cells. Cytotoxicity of *Morinda citrifolia* against mock-infected MT-4 cells was also assessed by the MTT method. Ether extract (EMC) of *Morinda citrifolia* exhibited a maximum protection of 58 % against replication of HIV-1 in MT-4 cells (EC_{50} =38 μ g/mL and CC_{50} = 66 μ g/mL). Methanolic extract (MMC) displayed cytostatic activity in MT-4 cells (C-type Adult T cell leukemia cells) with CC_{50} = 51 μ g/mL.

Key word: *Morinda citrifolia*, Anti-HIV activity, Cytotoxicity, MTT assay.

INTRODUCTION

Morinda citrifolia L (Noni) is a versatile medicinal plant with a broad spectrum of pharmacological activities. *Morinda citrifolia* is reported to possess hepatoprotective^{1,2} anticancer³, immunomodulator⁴, anti-inflammatory⁵, wound healing⁶, antioxidant⁷, anti-tubercular⁸ and wide spectrum of biological activity⁹ and safe herbal drug¹⁰. Recently, much attention has been devoted for searching potential antimicrobial agents from natural products, but anti-HIV activity of *morinda citrifolia* is relatively less explored. The present work is to study the anti-HIV activity of various extracts of the leaf powder of *morinda citrifolia* against HIV 1 and 2 in MT-4 cells.

EXPERIMENTAL

Materials and methods

The leaf powder of *morinda citrifolia* are dried under shade and powdered. The powder is extracted with different solvents (acetone, chloroform, methanol, ether and ethyl

* Author for correspondence; E-mail: periyasamy_selvam@yahoo.co.in

acetate) for five days by cold maceration. It is then filtered to get the extracts evaporated to dryness under vacuum. The dried extracts acetone (AMC) chloroform (CMC), ether (EMC), ethylacetate (EAMC) and methanol (MMC) were used for anti-HIV activity and cytotoxicity studies.

Anti HIV assay

Morinda citrifolia L. was tested for its inhibitory effects against replication of HIV-1 (IIIB) in MT-4 cells¹¹. The MT-4 cells were grown and maintained in RPMI 1640 DM Medium supplemented with 10% (v/v) heat-inactivated Fetal Calf Serum (FCS), 2 mM-glutamine, 0.1% sodium bicarbonate and 20 µg/mL gentamicin (culture medium). Inhibitory effect of *Morinda citrifolia* on HIV-1 replications was monitored by inhibition of virus-induced cytopathic effect in MT-4 cells and were estimated by MTT assay. Briefly, 50 µL of HIV-1 (IIIB) (100-300 CCID₅₀) were added to a flat-bottomed microtiter tray with 50 µL of medium containing various concentrations of *Morinda citrifolia*. MT-4 Cells were added at a final concentration of 6 x 10⁵ cells/mL. After 5 days of incubation at 37°C, the number of viable cells were determined by the 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide (MTT) method. Cytotoxicity of *Morinda citrifolia* against mock-infected MT-4 cells was also assessed by the MTT method. The anti-HIV activity and cytotoxicity data of *Morinda citrifolia* are presented in Table 1.

RESULTS AND DISCUSSION

Various extract of leaf powder of *Morinda citrifolia*. L has been evaluated for its anti-HIV activity and cytotoxicity (Table 1) against HIV-1 and -2 replication in acutely infected MT-4 cells. *Morinda citrifolia* extracts exhibited a maximum protection of 8-58 against the replication HIV-1 and -2 in acutely infected MT-4 cells. Ether extract of *Morinda citrifolia* (EMC) exhibited antiviral activity with EC₅₀=38 µg/mL and CC₅₀=66 µg/mL and 58% maximum protection against the replication HIV-1 in acutely infected MT-4 cells. Methanolic extract of *Morinda citrifolia* (MMC) displayed cytostatic activity against MT-4 cells (Adult T Cell leukemia) with CC₅₀= 51.20 ± 15.74 µg/mL.

The Polynesians utilized the whole Noni plant in various combinations for herbal remedies^{12,13} such as arthritis, diabetes, high blood pressure, muscle aches and pains, menstrual difficulties, headaches, heart disease, AIDS, cancers, gastric ulcers, sprains, mental depression, senility, poor digestion, atherosclerosis, blood vessel problems, and drug addiction. From this study, it has been observed that different extracts of leaf portion of *Morinda citrifolia* exhibits cytotoxic activity in MT-4 cells and ether extract (EMC) inhibits

the replication of HIV 1 below the cytotoxic concentration and further studies regarding isolation of active constituents from ether extract is under way.

Table 1: Anti-HIV activity of leaf extracts of *morinda citrifolia*

Extract	Strain	EC ₅₀ (µg/mL)	CC ₅₀ (µg/mL)	Maximum protection (%)
AMC	IIB	>138.75	138.75 ± 2.06	10
	ROD	>138.75	138.75 ± 2.06	8
CMC	IIB	>119.28	119.28 ± 18.35	11
	ROD	>119.28	119.28 ± 18.35	9
EAMC	IIB	>147	147 ± 19.71	10
	ROD	>147	147 ± 19.71	8
EMC	IIB	38.50	66 ± 0.06	58
	ROD	>49.70	49.70 ± 14.59	18
MMC	IIB	>51.20	51.20 ± 15.74	10
	ROD	>51.20	51.20 ± 15.74	15
AZT	IIB	0.00012	65.90	126
	ROD	0.00062	65.90	148

^a50% Effective concentration of compound, achieving 50% protection of MT-4 cells against the cytopathic effect of HIV.

^b50% Cytotoxic concentration of compound, required to reduce the viability of mock-infected MT-4 cells by 50%

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