INTRODUCTION

Since antiretroviral therapy has emerged as main tools against Human Immunodeficiency Virus (HIV), the prevalence and incidence of HIV were significantly reduced except for several countries in Europe and Central Asia. Several compounds from herbal extract have been reported to have an anti-HIV effect such as alkaloids, lignans. Those compounds attack the replicative cycle of HIV. Indonesia has widely known to have varieties of natural resources. One example of those plants that garnered attention to have anti -HIV is coming from the Moraceae family. The previous study stated that Moraceae family contain three first compound (from flavonoid) that showed anti-HIV, mulberryin, morusin and sanggenol. Other Ficus species such as Ficus glomerate extracted from wood (ethanol extraction) also showed a potent anti-HIV with IC50 7.8 µg/m.

OBJECTIVES

This study wants to evaluate the potential of other Ficus namely Ficus fistulosa which found in the tropical area including Indonesia as anti-HIV.

MATERIAL AND METHODS

Ficus fistulosa was extracted using ethanol as a solvent and further gradually fractionated in chloroform, ethyl acetate, and butanol solvents. The targeted persistently infected virus (MT4/HIV) cell lines were co-cultured with the herbal extracts at different time points and the syncytia and cytotoxicity assays were performed to evaluate the potential antiviral activity of Ficus fistulosa.

Cytotoxicity assay: Cell viability was quantified using the colorimetric WST-1 assay Cell Proliferation Reagent (Promega, USA). The viable cells were measured by the absorbance of each sample using a microplate reader at a wavelength of 450 nm. Cytotoxicity effect of Cdots and CBBA-Cdots were also evaluated on half of cytotoxic concentration (CC50) analyzed with Origin software.

Antiviral assay (syncytia formation):
Syncytia formation assay evaluated the antiviral activity of the as-prepared extract/fractions samples, amount of produced syncytia were microscopically counted. Inhibiting activity was also evaluated on half concentration of inhibition (IC50) value counted by Origin software.

CONCLUSIONS

Chloroform fractions of Ficus fistulosa showed antiviral activity against MT4/HIV cells. Though our study showed potential inhibition by chloroform fractions but further studies are required to investigate the active compound responsible for HIV inhibition in Ficus fistulosa extracts and fractions.

ACKNOWLEDGEMENT

The author wanted to thank all colleague in Universitas Airlangga Hospital for their effort in helping PBMC collection for this study and all member of Natural Product Research and Development Group from Institute of Tropical Disease Universitas Airlangga for helping in providing the Ficus fistulosa extract and fraction.

REFERENCES


RESULTS

Figure 1. Microscopic images consisting mix of cultured MOLT-4 cells and extract/fractions as positive control (no syncytia formation) (A), mix of cultured MOLT-4 cells and MT4/HIV-1 as negative control (B).

Evaluation of anti-HIV efficacy in vitro: In this study, we examined the HIV inhibitory activity of extract/fractions samples by setting the infection on cell-to-cell way and counting the numbers of syncytia produced after a 24-h incubation of cultured MT4/HIV-1 and MOLT-4 cells. Basically, syncytial formation is not equal to cell-to-cell pathway, but it represents the interaction between gp120 and CD4/CXCR4 during the pathway. Therefore, syncytia formation assay is a useful method for the screening of inhibitors for viral cell-to-cell transmission as shown in figure 1. We further analyzed the potency of extract/fractions samples on the inhibition of HIV infection by varying the concentrations of samples (Figure 2).

Figure 2. Inhibition intensity of extract/fractions samples against MT-4/HIV-1 cells over 24 h incubation compared negative control

Cytotoxicity assay: In order to evaluate the potential utility Ethanol Extract, Chloroform, Ethyl Acetate, and Butanol Fractions from Ficus fistulosa in biological applications, we assessed the toxicity of the obtained it in MOLT-4 human leukemia cells using WST-1 assays, as shown in Figure 3.

Figure 3. Cell viability evaluation using WST-1 assay of MOLT-4 cells after 24 h treatment with extract/fraction samples compared negative control

Two of the four tested extract/fraction showed antiviral activity against HIV. The crude extract showed effective inhibition as well as low level of toxicity (IC50= 8.50 µg/ml, CC50 = 42.35 ug/ml and SI =5.05). Meanwhile, Chloroform fraction also effectively inhibited the MT4/HIV cells proliferation while keeping the toxicity to a minimal level (IC50= 10.68 ug/ml, CC50 = 29.73 ug/ml and SI =2.78).