# CHAPTER I

# INTRODUCTION

## 1.1 Background

The worldwide dissemination of Human Immunodeficiency Virus (HIV) over the past four decades is one of the most catastrophic examples of the emergence, transmission, and propagation of a microbial genome.[1] Since the first cases of Acquired Immunodeficiency Syndrome (AIDS) were reported in 1981, infection with HIV has grown to pandemic proportion, UNAIDS estimates that there were 35.3 million (32.2-38.8 million) people living with HIV at the end of 2012. New HIV infection in 2012 were 2.3 million (1.9-2.7 million), and death due to AIDS in 2012 were 1.6 million (1.4-1.9 million).[2]

The AIDS epidemic in Indonesia is one of the fastest growing in ASIA. Indonesia, a country with a population of 237.5 million in 2010 has an estimated HIV prevalence of 0.27% among the 15-49 years age group. The cumulative number of reported HIV infections in Indonesia has risen sharply from 7,195 in 2006 to 127,416 by December 2013. The Ministry of Health estimates that without increased efforts to expand and strengthen prevention, treatment, care and support services across the country, Indonesia will have almost twice the number of people living with HIV and AIDS in 2014 as compared to 2008, rising from an estimated 227,700 to 501,400.[3-5]

Globally, the annual number of people newly infected with HIV continues to decline, this decrease is off set by the reduction in AIDS-related deaths due to the significant scale up of antiretroviral therapy (ART) over the past few years, declines in the annual number of AIDS-related deaths ilustrated the powerful health benefits of scaled-up antiretroviral treatment.[2, 6] In 2012, 9.7 million people in low-and middle-

income countries received antiretroviral therapy, representing 61% of all who were eligible under the 2010 World Health Organization (WHO) HIV treatment guideline.[2] In Indonesia, by December 2013, 39,418 people were receiving ART regularly from 418 sites across the country. Of ARV patients, 37,820 people (97%) were on 1st line ARV, and 1,186 people (3%) had been switched from first to second line ARV.[7]

The hallmark of HIV infection is the progressive loss or depletion of $CD4^+$ T lymphocytes.[8-13] The enumeration of peripheral $CD4^+$ T lymphocytes counts in HIV patients is essential for HIV staging, clinical management; taking a decision on initiation of ART, initiation chemoprophylaxis against opportunistic infection, a tool for monitoring disease progression, and the effectiveness of antiretroviral treatment. The changes in the $CD4^+$ T lymphocytes counts are important indicators of the response to antiretroviral treatment. [14-18] Actually, HIV plasma viral load is a sensitive indicator of the progression of HIV disease. However, due to relatively cost of viral load estimation, the $CD4^+$ T lymphocytes count remains the most important key indicator for initiation and monitoring of ART and a measure of the effectiveness of the treatment.[14, 15]

Key role of CD4+ T cells is to facilitate immune responses through production of immunomodulatory cytokines, the loss of these cells and the failure of remaining cells to function properly constitutes a critical impairment in immune capability.[19] The importance of $CD4^+$ counts as a strong predictor of opportunistic infections and non-AIDS events has been widely investigated, but little attention has been paid to the prognostic significance of $CD8^+$ counts. During untreated HIV infection, $CD8^+$ counts increase as $CD4^+$ counts decline.[20] a low CD4/CD8 ratio in elderly HIV-unifected adults is associated with increased morbidity and mortality. A subset of HIV infected adults

receiving effective antiretroviral therapy (ART) fails to normalize this ratio, even after they achieve normal $CD4^+$ T cell counts.[20]

Functional impairment of HIV-specific CD8 T-cells is a key indicator of immune status and disease progression in the HIV-infected individual. Cytotoxic T-lymphocytes (CTL) play an important role in the immune system's defense against HIV infection. CTL response is important in protection from HIV-infection. The regulation of HIV level is mostly mediated through specific CTL responses. Longitudinal studies during acute HIV-infection and highly active antiretroviral therapy (HAART) have demonstrated that viremia was decrease in association with increasing cytotoxic HIV-specific, $CD8^+$ T lymphocytes and HIV-specific $CD4^+$ T lymphocytes. Accurate assesment of the functional status of $CD8^+$ T cells is an important tool for managing the clinical therapy of HIV-infected patients.[21]

Cytotoxic T lymphocyte ($CD8^+$) cell activation is a major cause of HIV pathology. The expression of the activation markers CD38 and HLA-DR on T cells is an indicator of cell activation.[22] CD38 is a membrane-bound adenosine 5'-diphosphate (ADP) ribosyl cyclase. These marker of activation are elevated in direct proportion to the magnitude of HIV replication and some studies have found that the extent of CD38 expression predicts ultimate HIV disease course more accurately than plasma levels of HIV.[23] $CD8^+$ cellular immune activation surface markers ($CD8^+$/HLA-DR/CD38) as opposed to $CD4^+$ activation, is more predictive of long-term immunologic responses.[24] The analysis of CD38 expression on lymphocytes has become an important tool for monitoring patients during HIV-1 infection and has recently been proposed for use in the follow-up of highly active antiretroviral therapy (HAART).[25-27] An observational study of children vertically infected with HIV-1 was performed to determined the role of CD38 expression in $CD8^+$ T cells as prognostic marker of virological failure in

children receiving HAART. The $CD8^+CD38^+$ T cell count not only predicts progression of HIV disease to AIDS and death, but it also offers additional independent predictive value for evaluation of plasma viral load (VL) and $CD4^+$ T cell count.[28]

Humans infected with HIV have been shown to be under chronic oxidative stress. Oxidant production could enhance HIV replication via activation of nuclear factor-κB ($NF_KB$) and indirectly through activation genes that further promotes oxidative stress and hence HIV replication.[29-31] Interestingly, HAART proved to have deleterious effects as a result of mitochondrial dysfunction, increased in oxidative status and it plays an important role in the occurence of oxidative stress.[32, 33] Antioxidant, may have a role in the treatment of HIV infection.[31, 34] Mangosteen (*Garcinia mangostana*) is one of the tropical fruit that the extract of various parts contain varieties of secondary metabolites such as prenylated and oxygenated xanthones.[35, 36] Experimental studies have shown that obtained xanthones from mangosteen have remarkable biological activities as antioxidant, antitumor, antiallergic, antiinflammatory, antibacterial, antiviral activities,[35-39] antiplasmodial, cytotoxic and potensial cancer chemoprotective activities.[36] Moreover, some of the xanthones from mangosteen have been found to influence specific enzyme activities, such as aromaterase, inhibitor kB kinase, quinone reductase, sphingomyelinase, topoisomerase and several protein kinases.[36] Several studies showed that xanthones from Garcinia mangostana act as an active constituents against HIV-1 protease.[40-42] A randomized controlled trial study in healthy adults showed that intake of a xanthone-rich mangosteen product for 30 days increases in peripheral $CD4^+/CD8^+$ T cell frequency. The result indicated that the intake of an atioxidant-rich product siginificantly enhanced immune responses and improved the subject's self-appraisal on overall health status.[43]

Although it has been many research related to Garcinia mangostana performed, research on the effects of mangosteen (*Garcinia mangostana)* peel extract against $CD8^+$ T lymhocytes and CD38 expression in HIV patients has not been investigated, thus not known whether mangosteen *(Garcinia mangostana)* peel extract effect on the CD8+ lymphocytes and CD38 expression in HIV patients with HAART.

## 1.2 Research Question

Does mangosteen *(Garcinia mangostana)* peel extract has effect on $CD8^+$ T lymphocytes and CD38 expression in HIV patients with antiretroviral therapy ?

## 1.3 Research Objective

### 1.3.1 General Objective

To determine the effects of mangosteen *(Garcinia mangostana)* peel extract towards $CD8^+$ T lymphocytes and CD38 expression in HIV patients with antiretroviral therapy.

### 1.3.2 Specific Objective

1. To determine differences in number of $CD8^+$ T lymphocytes in HIV patients with antiretroviral therapy given mangosteen *(Garcinia mangostana)* peel extract and control group.
2. To determine differences in the level of CD38 expression in HIV patients with antiretroviral therapy given mangosteen *(Garcinia mangostana)* peel extract and control group.

## 1.4 Benefit of Study

### 1.4.1 Benefit for science

The result of this study are expected to improve the knowledge about the use of mangosten (*Garcinia mangostana)* peel extract against several immunological parameter and relevant to the development of herbal research.

### 1.4.2 Benefit for patient management

By the result of this study, it will be known whether mangosteen (*Garcinia mangostana)* peel extract can be considered as an adjuvant treatment for HIV patients with antiretroviral therapy, mainly related to the number of $CD8^+$ T lymphocytes and the level of CD38 expression, where these two parameters can describe the prognosis of therapeutic efficacy and disease progression.

### 1.4.3 Benefit for further research

Result of this study can be used as consideration for further research in HIV patients.

## 1.5 Research Originality

**Table 1.1 Research originality**

| No. | Author, title of publication and journal | Methods | Results |
|---|---|---|---|
| 1. | Tang Y-P, Li P-G, Kondo M, Ji H-P, Kou Y, Ou B. Effect of a mangosteen dietary supplement on human immune function: a randomized, double-blind, placebo-controlled trial. *Journal of medicinal food*. 2009;12:755-63 | A randomized, double blinded, placebo-controlled study was conducted in 59 healthy human subjects (40-60 years old). Changes from baseline immune function were measured after a 30-day consumption of the | A xanthone-rich mangosteen product intake increased peripheral T-helper cell frequency, reduced the serum CRP concentration, increases in peripheral CD4/CD8 T-cell frequency, serum |

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| | | mangosteen product and the placebo | complement C3,C4, and concentrations IL-1alpha IL-1beta concentrations in the experimental group than in the placebo group |
| 2. | Palakawong C, Sophanodora P, Pisuchpen S, Phongpaichit S. Antioxidant and antimicrobial activities of crude extracts from mangosteen (Garcinia mangostana L.) parts and some essential oils. *International Food Research Journal*. 2010;17:583-9 | The antioxidant and antimicrobial activities of the extracts from peel, leaves, and bark of mangosteen (*Garcinia mangostana* L.), and some essential oils such as cinnamon and citrus were investigated. The antioxidant activities (IC50) of peel, leaves, and bark extracted, which were evaluated by DPPH method | These sample data received show the inhibition potency on extracts of peel, cinnamon, and the citrus oil that can be considered as preservative agents for both antibacterial and antioxidant activities. |
| 3. | Magadula JJ, Tewtrakul S. Anti-HIV-1 protease activities of crude extracts of some Garcinia species growing in Tanzania. *African Journal of Biotechnology*. 2010;9(12):1848-52 | Eighteenth ethanol extracts from some *Garcinia* species in the Guttiferae (Clusiaceae) family collected in Tanzania were investigated for their HIV-1 protease (HIV-1 PR) inhibitory activities using high performance liquid chromatography (HPLC) | The isolation of active compounds from These plants showed the most potent inhibitory activity against HIV-1 PR |
| 4. | Al-Massarani SM, El Gamal Aa, Al-Musayeib NM, Mothana Ra, Basudan Oa, Al-Rehaily AJ, et al. Phytochemical, antimicrobial and antiprotozoal evaluation of Garcinia mangostana pericarp and α-mangostin, its major xanthone derivative. *Molecules (Basel, Switzerland)*. 2013;18:10599-608. | Five xanthone derivatives and one flavanol were isolated from the dichloromethane extract of *Garcinia mangostana* and evaluated *in vitro* against protozoal, microbe and fungal. | α-mangostin showed high cytotoxicity and a broad but non-selective antiprotozoal and antimicrobial activity profile |

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| 5. | Tewtrakul S, Wattanapiromsakul C, Mahabusarakam W. Effects of compounds from Garcinia mangostana on inflammatory mediators in RAW264.7 macrophage cells. *Journal of Ethnopharmacology*. 2009;121:379-82 | The extract of *Garcinia mangostana* together were tested for anti-inflammatory effect against lipopolysaccharide (LPS)-induced nitric oxide (NO), prostaglandin E2 (PGE2), tumor necrosis factor alpha (TNF-α) and interleukin-4 (IL-4) releases aswell as their mechanisms in transcriptional levels using RAW264.7 macrophage cells | Mangosteen extract possessed potent NO inhibitory effect, PGE2 release and possessed only moderate effects towards TNF-α and IL-4 releases.Both extract and α-mangostin suppressed transcription of gene encoding inducible nitric oxide synthase (iNOS) and cyclooxygenase-2 (COX-2) in dose-dependent manners, whereas _-mangostin had only an inhibitory effect on transcription of iNOS. |

This current study differs from previous studies because it had never performed research on the effect of mangosteen *(Garcinia mangostana)* peel extract towards $CD8^+$ T lymphocytes and CD38 expression in HIV patients with antiretroviral therapy.