ICAM-1 OVEREXPRESSION INDUCED BY ABACAVIR IS MEDIATED BY P2X7 RECEPTORS
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INTRODUCTION
The use of abacavir has been associated with cardiovascular diseases. Specifically, it has been observed an increased risk of myocardial infarction (MI), arteriosclerosis and thrombosis in abacavir treated patients (1,2), but the mechanism remains unclear. In earlier studies, we have demonstrated that abacavir induces an increase in endothelial intercellular adhesion molecule-1 (ICAM-1) expression and promotes leukocyte recruitment through Mice-ICAM-1 interaction (2-4). Given the chemical structure of abacavir, we have previously explored the link between abacavir and its proinflammatory effects by interfering in the purine signalling pathway, and have seen that ATP and its P2X7 receptors (oxATP and A804598) and the agent responsible for ATP hydrolisis (Apyrase), but not the selective antagonist of P2X2/3 receptors (A317491) prevented ICAM-1 overexpression on HUVEC and on HUAEC. Abacavir produced a significant increase on leukocyte adhesion on HUVEC. A non-selective antagonist (Suramin), selective antagonist of P2X7 receptors (oxATP and A804598) and the agent responsible for ATP hydrolisis (Apyrase) prevented the increase on leukocyte adhesion.

AIM
The aim of the present study was to evaluate the role of ATP and its receptors on the endothelial ICAM-1 overexpression induced by abacavir.

RESULTS
Flow Cytometry
The expression of the endothelial adhesion molecule ICAM-1 was evaluated using an FACScalibur cytometer (BD Biosciences). ICAM-1 was analyzed in HUVEC/HUAEC incubated with the anti-ICAM-1 antibody conjugated with phycoerythrin (PE). The expression of the endothelial adhesion molecule ICAM-1 was evaluated using an FACScalibur cytometer (BD Biosciences). ICAM-1 was analyzed in HUVEC/HUAEC incubated with the anti-ICAM-1 antibody conjugated with phycoerythrin (PE).

Figure 1. Role of ATP receptors on leukocyte accumulation induced by abacavir on HUVEC.

Figure 2. Representation of the binding/fixation of ICAM-1 expression on HUVEC.

Figure 3. Role of ATP receptors on ICAM-1 expression-induced by abacavir on HUVEC.

Figure 4. Role of ATP receptors on ICAM-1 expression-induced by abacavir on HUAEC.

Abacavir promoted a significant increase on endothelial adhesion molecule ICAM-1 on HUVEC (Fig 3) and on HUAEC (Fig 4). A non-selective antagonist of ATP receptors (Suramin), the selective antagonist of P2X7 receptors (oxATP and A804598) and the agent responsible for ATP hydrolisis (Apyrase), but not the selective antagonist of P2X2/3 receptors (A317491) prevented ICAM-1 overexpression on HUVEC and on HUAEC. Significant *p<0.05 (vs. control) and +p<0.05 (vs. abacavir), respectively.

CONCLUSIONS
Our results suggest that the activation of P2X7 receptors promote overexpression of ICAM-1 in the venular and arterial endothelium. This process may be responsible for the leukocyte recruitment observed in the vascular damage associated with arteriosclerosis and myocardial infarction in HIV patients treated with abacavir.

REFERENCES

FLOW CYTOMETRY
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EXPERIMENTAL PROTOCOL
To study the effects of blocking P2X7 receptors on ICAM-1 expression, human umbilical vein or arterial endothelial cells (HUVEC or HUAEC, respectively) were pre-treated with the following reagents:

- Suramin (1 UI/ml ATP Hydrolisis - APYRASE
- oxATP (1 µM P2X7 antagonist - A317491)
- A804598 (50 µM P2X7 antagonist - oxATP)

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