Rosario Rossi; Anna Chiara Nuzzo; Giovanni Guaraldi*; Antonella Lattanzi; Maria Grazia Modena

Study of endothelial function in patients with HIV infection. What adds to the clinical evaluation? How and why should such patients be studied?

Institute of Cardiology, Policlinico Hospital, University of Modena and Reggio Emilia
*Institute of Infectious Diseases, Policlinico Hospital, University of Modena and Reggio Emilia

Corresponding author: Rosario Rossi  MD
Institute of Cardiology. Policlinico Hospital
Via del Pozzo 71, 41100 Modena (MO) Italy
Tel.: +39 059 4225505 - Fax: +39 59 4223714 - E-mail: rossi.rosario@unimore.it

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Introduction

In 2006 forty million people worldwide were living with human immunodeficiency virus (HIV), of whom 4.9 million were newly infected, with half of these infections occurring in individuals younger than 25 years. Combined antiretroviral therapy (cART) has greatly reduced the risk of potentially fatal opportunistic infections and thus has extended the life-span of these patients. Therefore, other possible causes of morbidity and mortality in the HIV-positive population have come to the forefront. In recent years, several clinical studies have suggested that HIV-infected patients experience high rates of cardiovascular disease. Clearly, acute coronary syndromes, namely myocardial infarction, are among these potential causes of morbidity and mortality in the HIV-positive population.

Patients with HIV infection are patients with high cardiovascular risk, as they are prone to developing more cardiovascular events than their peers of the same sex and ethnic group (1). Cardiovascular events in general and coronary events in particular are characterized by a high mortality rate, which can reach up to 25-30%. In addition, approximately one quarter of cardiovascular events occur without premonitory clinical and instrumental signs. For these reasons, the cardiovascular risk of each patient should be carefully stratified during the initial clinical visit, i.e. before the first event occurs! This serves to assist clinicians in deciding the type and intensity of treatment.

The assessment of cardiovascular risk in patients with HIV, like all patients, should be a multi-factorial evaluation, taking into account several determinants. At one time it was thought that the risk of having a myocardial infarction depended only on the presence of a significant number of coronary atherosclerotic plaques. While this remains largely true today, the risk is, in addition, attributed to other important factors, such as a tendency towards hypercoagulation (the old "thrombophilic diathesis"), and a "scarring" myocardium that is prone to develop arrhythmic events which could lead to sudden death. This theory is known as "theory of the vulnerable patient", masterfully set out in 2003 by a task force of experts belonging to the American Heart Association (2). This theory, universally accepted by the cardiological world, focuses on the "overall" cardiovascular risk, i.e., on the individual patient's "global" risk of developing events.

Most clinicians have no problems when it comes to determining which patients are in a situation of high cardiovascular risk. They know these patients are mainly those who have already suffered a cardiac event (acute myocardial infarction, or stroke, for example); or those who already have clinical manifestations of atherosclerotic disease, or have atherosclerotic plaques disclosed by imaging (for example, a carotid plaque demonstrated by ultrasound), or patients with diabetes mellitus, or, finally, patients with multiple (three or more) "conventional" cardiovascular risk factors (hypertension, hypercholesterolemia, cigarette smoking, advanced age, obesity). Irrespective of HIV infection, treatment in these patients must be aggressive and conducted in ways that "optimize" treatment (reaching a target blood pressure <120/80 mmHg, an LDL cholesterol <100 mg/dL, physical activity for one hour at least three times a week, losing excess weight, and so on).

Moreover, it is not difficult to classify patients at low cardiovascular risk. They are those who have no risk factors, or they have one, that is well controlled by medication. The right choice in these patients is to re-evaluate with regular follow-up. What is certainly the most difficult is the proper management of the patient at intermediate risk, either because the patients in this category of risk are the majority (55-60%), both because it is not clear in these subjects which therapeutic strategy will reduce the risk. Patients at intermediate risk are those who have two or three risk factors, often not adequately controlled by medication. How should these patients be treated? It is very difficult to answer this question properly because no studies have addressed this category of subjects. For this reason we must seek to reclassify, establishing who truly deserves an aggressive strategy and who, on the contrary, can be regularly followed up, in order to optimize resources. At this point, physicians should provide a tool that enables them to make...
a more precise classification and a more accurate assessment of cardiovascular risk. In this light we propose the study of endothelial function. A first answer is therefore clear. Should endothelial function be studied in such patients? Certainly not in the high-risk patients, in whom treatment should in any case be aggressive; nor in the patients at low risk, for whom an attitude of “wait and see” must prevail; but certainly in patients at intermediate risk, in order to better classify them. Only in these patients do we believe the study of endothelial function can add something from clinical practice.

**Why and how should endothelial function be studied?**

The endothelium is a dynamic organ system which maintains normal vessel homeostasis by regulating vascular tone and protecting against atherogenesis. Impairment of endothelial function is not only an important initial step in the pathogenesis of atherosclerosis, but is also associated with a defective endothelial repair mechanism which may lead to further progression of vascular disease. The so-called “response-to-injury theory” postulates that endothelial dysfunction is the product of all cardiovascular risk factors. This malfunctioning would determine the initiation, growth, and also complication of atherosclerotic plaque, and this can lead to an acute cardiovascular event. Endothelial dysfunction is, therefore, the most precocious phenomenon linked to atheroma formation. It follows that a normal-functioning endothelium is not compatible with the atherosclerotic disease (3). There is a double-wire link between HIV infection, endothelial dysfunction and atherosclerosis. The close relationship between the virus and atherosclerosis is that HIV is able to infect endothelial cells. In vitro infection of human umbilical vein endothelial cells with HIV-1 has been demonstrated (4). There is also evidence that the infection of endothelial cells is “productive”, i.e. HIV-1 DNA was detected in brain microvascular endothelial cells of HIV-1-positive patients (4,5). This phenomenon probably affects the malfunctioning of the endothelial cells. In fact, cART-naive patients display markers of endothelial activation. Plasma levels of von Willebrand factor, plasminogen activator inhibitor-1 antigen, and tissue-type plasminogen activator are significantly elevated in HIV-1-positive patients (6,7). Compared with healthy controls, antiretroviral-naive HIV-1 positive subjects have higher levels of soluble vascular cell adhesion molecule-1 (8) intracellular adhesion molecule-1, and E-selectin (9,10). This up-regulation of cell adhesion markers in cART-naive patients suggests that the virus activates and dysregulates endothelial cells.

Elucidation of the important role of the endothelium in various stages of atherosclerosis has led to the development of tests designed to evaluate the functional properties of the vascular endothelium. Since 1992, the noninvasive measurement of endothelium-dependent, brachial artery flow-mediated dilatation (FMD) has been used by numerous investigators to evaluate endothelial function (11). Using high-resolution ultrasound, this method quantifies the change in diameter of a conduit artery (usually the brachial artery) in response to shear stress induced by increasing blood flow during reactive hyperemia. Mounting evidence suggests that brachial FMD is not only associated with traditional cardiovascular risk factors but also with future cardiovascular events (12-17). Several papers have now assessed HIV infected patients using FMD to better evaluate global cardiovascular risk. The majority of studies highlighted the direct link between the virus and the endothelium. Higher HIV viral load was in fact inversely correlated with reduced endothelium-dependent FMD in a controlled case-study of a group of HIV positive individuals, independent of cART regimens (18). Solages et al. (19) monitored FMD in 75 HIV infected patients and 223 control subjects, and found significantly impaired endothelial function in the infected population. Viral load was a significant predictor of reduced FMD. A study in patients aged from 3.5 to 19.5 years also showed that HIV-infected children had significantly lower FMD than noninfected children or cART-naive children (20). In a recent study, Oliviero et al. (21) demonstrated that HIV per se is able to stimulate atherogenic processes inducing endothelial dysfunction. In that study brachial artery FMD was impaired in HIV patients and was significantly more impaired in the subgroup of patients with viral load values above the median. In addition, there was a highly significant inverse correlation between FMD and the HIV-RNA copies. Another important multicenter study demonstrated how HIV had in a deleterious effect on endothelial cells (22). Treatment with cART (the author tested three different potent regimens) led to an increase of CD4 cell count and a decrease of HIV RNA copies, and this was accompanied by a significant improvement of endothelial function.

**Why is the study of endothelial function by FMD evaluation not used in all centers?**

The evaluation of FMD is non-invasive, non-bloody, may be repeated, and can also be done at the patient’s bedside, but still presents some limitations that must be taken into account. Firstly, methodologies used for the measurement of FMD are not standardized among various vascular laboratories. Thus, there are no universal cut-off values for FMD, making it difficult to compare the results among different laboratories. Secondly, there is a wide variation in intersession FMD likely due to physiologic fluctuations. Thus, single measurements of FMD may have limited utility. Thirdly, high quality ultrasound systems, capable of quantifying a hyperemic response of 0.2 to 0.3 mm difference in a brachial artery, are necessary for reproducibility and validity of the technique. Thus, high cost and technical difficulties prevent widescale clinical application of FMD evaluation. Lastly, the lack of an accepted diagnostic gold standard for endothelial function hampers evaluation of the diagnostic accuracy of brachial artery FMD.

Notwithstanding these limitations, the consistent results obtained in many studies suggest that an impaired brachial FMD is associated with future cardiovascular events beyond conventional cardiovascular risk factors across a wide variety of populations, also in HIV infected patients.
REFERENCES


