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Shehata's Test for Organ Donation and Transplantation

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Abstract

While the genetic matching between the donor and the recipient is essential for the success of the transplant procedure, there are other genetic factors that have the potential to significantly influence the clinical outcome. In this paper, a relatively simple test is described that has the potential to predict the suitability of the potential donor for organ donation. This test should be performed by the time the person makes his decision to be a future donor.

Keywords: IL6; IL10; Genetic polymorphisms/mutations; Solid organ transplants

Because of the increasing incidence of the chronic diseases of the vital organs, such as lung, heart, liver and kidneys, the importance and frequency of the solid organ transplantation procedures are increasing [1]. At the moment, there are many patients on the waiting lists for transplantation, which is further complicated by the incidence of post-transplantation organ dysfunction and or failure [2] that lead to high morbidity and mortality.

State of the Art

According to many studies, the ratio between interleukin (IL) 6 and 10 plays an important role in the modulation of the immune response, which determines the success of the transplantation procedure.

Interleukin-6 (IL-6) is a proinflammatory cytokin and a proinflammatory myokin. In humans, it is coded by the IL-6 gene. IL-6 is secreted mainly by the T-cells and macrophages of the graft, bone marrow, spleen, lymph nodes, brain and skin. The nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB) is the main regulator of IL-6 expression, which increases in all cases of inflammation and tissue injuries [3,4].

IL-6 is secreted by the vascular smooth muscles as a proinflammatory cytokine, however, it can work indirectly as an anti-inflammatory cytokine through the antagonization of TNF-alpha and IL-1, as well as the activation of IL-10 [5].

IL-10 is a cytokine with anti-inflammatory activities. It regulates the expression of the cytokines in the T-helper 1 cells and the expression of the major histocompitability complex class-II antigens and the other co-stimulatory molecules on the surface of macrophages. Moreover, IL-10 antagonizes the activities of NF-κB [6].

Accordingly, the balance between IL-6 and IL-10 has a great potential to affect the prognosis of many inflammatory conditions, including the ischemic-reperfusion injury (IRI) and the graft-host interaction. Thus, the ratio between both cytokines has the potential to predict the prognosis of the transplantation procedure and the occurance of the graft dysfunction/failure after transplantation [5]. A high IL-6/IL-10 ratio after transplantation is associated with primary graft dysfunktion and 20-fold increased relative risk of mortality [7].

The Problem and the Solution

Based on the above discussed principles, many studies and transplantation centers use the ratio between IL-6 and IL-10 as a marker for the transplantation prognosis. However, till now, the levels of those cytokines are measured either in the ex vivo perfusate of the graft (representing the graft levels after death of the donor and the experience of IRI) or following transplantation (the levels in the recipient's blood), which doesn't save the precious time and costs.

As the main source of the graft cytokines is the resident leucocytes, a prior assessment of the mutations/polymorphisms associated with increased production/function of IL-6 and or decreased production/function of IL-10 (in addition to any other relevant genetic markers of inflammation) in the leucocytes of the donor can provide valuable information about the suitability of the potential donor.

Accordingly, clinical studies should be conducted to evaluate the genetic profile of the genes of interest before donation, and the evidence-based association with the success rates of the transplantation procedures, as well as the frequencies of complications or graft failure following transplantation. This could be of great value in order to, at least, mark the future grafts as a "relatively high risk", which would indicate the application of special immunomodulatory interventions, such as *ex vivo* organ perfusion.

Shehata's Test

A simple strip assay for detecting the genotypes of the IL-6 and IL-10 genes in the leucocytes of a blood sample should be performed during the decision-making for being a future organ donor, in order to assess whether the future grafts would have an increased IL-6/IL-10 ratio after death and IRI. The technique consists of the following steps:

A blood sample is to be collected

- Leukocytes are to be separated by a suitable protocol or technique.
- DNA from leukocytes is to be extracted.
- PCR reaction and simultaneous labeling by fluorescent-labeled highly-specific primers for all known IL-6 and IL-10 mutations and polymorphisms, provided that each mutation or polymorphism has a specific fluorescence color.
- PCR products are then to be hybridized to the test strip.
- The test strip has the various known genotypic sequences of IL-6 and IL-10, which are highly complementary to the PCR products, fixed on it.

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- The fixed genotypes on the test strip are colorless.
- When the fluorescent-labeled PCR products hybridize with the complementary genotypes, the others are washed out, so that, only the fixed colors can be seen and identified on the test strip under the fluorescent microscope.

Further Readings

Mohamed MSA (2016) Role of Genetic Testing in Lung Transplantation; Prediction of Inflammation. J Genet Syndr Gene Ther 7: 298. doi:10.4172/2157-7412.1000298.

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