Cytokine response in severe sepsis — Predicting and modelling the course of illness

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Background: Severe sepsis remains one of the most threatening conditions in intensive care. During the progression of sepsis from early hit to multiorgan failure proinflammatory and anti-inflammatory cytokines are released. Cytokines can be used as a biomarkers to determine the specific patterns of sepsis progression and association with mortality (1). These biomarkers were successfully used as predictors in animal studies (2). Data from humans, especially comparison between children and adults, are limited. Hence, in this study we widely describe systemic cytokine response in this type of patient population.

Methods: Prospective study of 37 subjects (20 children, 17 adults) hospitalized with severe sepsis in intensive care. We measured CRP, procalcitonin, TNF, IL-1beta, IL-4, IL-6, IL-8, IL-10, IL-12, TREM-1. ANOVA models were specified using Proc Mixed. Study was fully approved by an EC.

Results: We identified a correlation of CRP levels with mortality or presence of shock. We found a distinct feature of CRP in adults with pronounced dynamic dichotomy in these subjects. Levels of IL-6 were significantly different in adult patients in the context of shock states. Highest risk of death was in adult patients associated with TREM-1 sustained high after 48 hours after sepsis onset. Otherwise, there was no correlation with death, shock states and SOFA score for PCT, TNF, IL-1beta, IL-4, IL-8, IL-10, and IL-12.

Conclusion: Response of circulating factors in patients with severe sepsis is heterogeneous in adults and children population and has some distinct features according to dynamics of CRP, IL-6 and TREM-1. We can find an evident discriminative feature of CRP and TREM-1 value dynamics. Sustained high levels of CRP and TREM-1 48 hours after sepsis onset were predictive of high risk of death. This finding could be related to the progression of illness to multiorgan failure. An activation of proinflammatory cytokines is associated with higher severity of sepsis and is probably related to severity of the initial hit. Supported in part by the Internal Grant Agency of the Ministry of Health NR 9297-3 and NR 9894-4.

Adults, Solid line: Exitus, Dashed line: survived.

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Circulating antigens of Mycobacterium tuberculosis: Standardization of immunoenzymatic assay to detect circulating antigens related in the pathology of tuberculosis

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Background: In 2006, an estimated 9.2 million new cases of tuberculosis in worldwide (WHO 2008). WHO has been set goals for global TB control including early detection of infected individuals and the prompt treatment. For this end, it has been encouraged public-private partnerships to find solutions that would drive to the development of alternative diagnostics methods. In attention to WHO recommendations immunological tests are currently being studied and standardized for the diagnosis of the active and latent disease. Based on our laboratory expertise, we have been worked to standardize immunoassays to detect Mycobacterium tuberculosis antigens. Objective: To standardize enzyme immunoassay to detect circulating antigens of M. tuberculosis in serum samples from patients with active pulmonary tuberculosis.

Methods: 6-kDa early secreted antigen target (ESAT-6), protein low molecular weight, secreted by M. tuberculosis and absent in BCG vaccine strain; 16kDa, heat shock protein, important in the persistence of the bacillus, antigen 85 complex is suggested to be involved in the binding of mycobacteria to specific surface receptors of macrophages, were the antigens studied. For this, plastic plates were coated with monoclonal antibodies anti-ESAT-6, 16kDa, Complex 85. In order to avoid false results we studied three block solutions (BSA 2%, 5% and 2% skim milk in PBS). Pool of normal human serum was spiked with different concentrations of proteins from H37Rv strain (500 μg/mL to 100 μg/mL) to determine the ELISA analytical sensitivity. The immune reaction was detected using a rabbit anti-M. tuberculosis - peroxidase diluted to 1/500.

Results: Better results were obtained with plates coated with monoclonal anti-ESAT-6 and blocked with 2% BSA in PBS.