Revista Brasileira de Hematologia e Hemoterapia
Brazilian Journal of Hematology and Hemotherapy
www.rbhh.org

Original article

Normal lymphocyte immunophenotype in an elderly population

Sâmia Macedo Queiroz Mota Castellão Tavares[a,*], Wilame de Lima Bravo Junior[b], Metton Ribeiro Lopes e Silva[b]

[a] Faculdade Leão Sampaio (FALS), Juazeiro do Norte, CE, Brazil
[b] Laboratório de Análises Clínicas Vicente Lemos, Crato, CE, Brazil

ARTICLE INFO

Article history:
Received 27 March 2013
Accepted 2 February 2014
Available online 4 April 2014

Keywords:
Elderly
Lymphocyte count
Immunophenotyping
Immunity
Disease

ABSTRACT

Objective: The aim of this work was to evaluate the lymphocyte immunophenotype in an elderly population.
Methods: This study enrolled 35 over 60-year-old volunteers and a control group composed of 35 young adults. The study included elderly without diseases that might affect the functioning of the immune system. These individuals were consulted by doctors and after a physical examination, laboratory tests were performed using a Beckman Coulter® flow cytometer. The GraphPad Prism computer program was employed for statistical analysis with the level of significance being set for p-values <0.05.
Results: There is a statistically significant reduction in the number of lymphocytes ($CD8^+$, $CD2^+$ and $CD3^+$ cells) in the elderly compared to young adults. These low rates are explained by changes attributed to aging and may be partly responsible for the reduction in the cellular immune response, lower proliferative activity and the low cytotoxicity of lymphocytes.
Conclusion: These parameters showed greater impairment of adaptive immunity in the elderly population and can therefore explain the greater fragility of the aged body to developing diseases.

Introduction

The immune system is our first line of defense against foreign agents. These invaders, viruses, bacteria and fungi, can be moderately aggressive, such as those responsible for the common cold or more harmful such as in meningitis or tuberculosis. However, this system has several internal control mechanisms that prevent the development of infections. The defense mechanisms range from mechanical protection to complex cellular and molecular mechanisms.[1]

Innate immunity acts as a first line of organism defense. The innate immune system consists of several components. First, the epithelial barrier prevents infections and if this defense is destroyed, a group of phagocytic cells are activated, including monocytes, macrophages and neutrophils.[2]

* *Corresponding author at*: Rua Dr. Miguel Lima Verde, 513, Centro, 63100-060 Crato, CE, Brazil.
E-mail address: novosamia@gmail.com (S.M.Q.M.C. Tavares).
http://dx.doi.org/10.1016/j.bjhh.2014.03.021

In addition, the immune system can elicit another type of response called the adaptive immune system, which is also effective but slower and longer lasting. Its main characteristics are immune memory and specificity. In acquired immune responses, lymphocytes, classified into two basic categories as T lymphocytes and B lymphocytes, act to destroy pathogens.[3]

With aging, the elderly are affected more by infections and are more likely to develop cancer than younger individuals. These clinical problems are attributed, at least in part, to the aging of the immune system, immunosenescence, which is associated with an imbalance in the immune function.[4]

One theoretical basis to explain the aging process is that the immune activity, similar to most other physiological functions, declines with age.[5] Conversely, studies have shown that there is an increase of natural killer (NK) cells with increasing age. The real cause of this increase remains unknown, but it is believed that this compensates for the decline in T-cell response.[6]

Neutrophils, important cells of innate immunity, do not respond efficiently in the mobilization process of the elderly when the hematopoietic system is under stress, for example, during chemotherapy and in severe prolonged infections. In these circumstances, the process of migration of neutrophils from the bone marrow into the systemic circulation is not as active as in youth people.[7] Moreover, studies with different age groups demonstrated a significant decline in the phagocytic ability of neutrophils against bacteria as well as in the number of bacteria engulfed with advancing age.[8]

Research has shown that aging is accompanied by progressive changes in the composition of lymphocyte subsets (CD4 and CD8) in lymphoid tissues, with changes associated to T-cell function including those in relation to cytokine secretion.[6]

One of the major changes in the immune response in respect to the senescence system is the reduction of virgin T cells and increase in memory clones, in other words, $CD4^+$ and $CD8^+$ memory T cells expand while $CD4^+$ and $CD8^+$ virgin T cells gradually decrease with aging.[9]

An evaluation of the immune system may help to develop interventions in the elderly to avoid health problems that would eventually affect the independence of these individuals. Thus, early treatment may revert damage that under other circumstances would become irreversible.[10]

Objective

The aim of this study was to evaluate the profile of the normal lymphocyte immunophenotypes in a population of healthy elderly people.

Methods

This research is a descriptive study of lymphocyte immunophenotyping in an elderly population. Therefore, participants should be at least 60 years old and as healthy as possible.

Interviewers visited more than 200 homes to ascertain whether any of the residents were over 60 years old. They explained the purpose of the research and invited those who were interested to participate in the study.

The study group was selected by the Geriatrics Department, University Hospital Walter Cantídio (UHWC) of the Universidade Federal do Ceará (UFCE). For the physical evaluation, physicians requested basic biochemical tests, including complete blood count, total cholesterol, triglycerides, blood sugar, and creatinine in order to better assess each individual. After this evaluation, the study enrolled 35 of 200 elderly, who were physically healthy and had normal results for all these exams.

Informed consent was obtained from all participants and the study was approved by the Research Ethics Committee of the UFCE.

Thus, the population consisted of 61- to 92-year-old individuals of both genders and without chronic diseases, such as rheumatoid arthritis and systemic lupus erythematosus. Moreover the participants did not have acute infections such as colds and viruses, mental disability or had been diagnosed with depressive syndrome.

Individuals with autoimmune diseases, or other problems that may impair the immune system and those suffering from renal failure, severe heart disease or anemia were excluded from the study. Other exclusion criteria were the use of corticosteroids or immunosuppressive medications.

A control group of randomly chosen young adults was also created mainly of companions of patients who were waiting to be consulted at HUWC. Controls were submitted to the same tests as the study group and only those with normal results were enrolled in the study.

Blood samples were collected by venipuncture using sterile disposable equipment in a tube with 5 mL of ethylenediaminetetraacetic acid (EDTA) as anticoagulant. This sample was used to perform immunophenotyping (markers of lymphocytes) with all tests being carried out on the same day the blood was drawn to guarantee the results.

The investigation of the immunophenotype was made using a Beckman Coulter® flow cytometer to obtain counts and percentages of the subpopulations of lymphocytes.

Following the manufacturer's instructions, 100 mL of whole blood were incubated for 15 minutes with a combination of labeled monoclonal antibodies (10 mL each) in the dark at room temperature. Monoclonal antibodies, of the same brand as the flow cytometer, were conjugated with specific fluorochromes. After incubation, the samples were subjected to lysis using a standard reagent (OptiLyse® C).

Cells with the different markers (CD3, CD4, CD8, CD2, CD19) were counted using a technique that has been described previously[11]. Data are reported as absolute numbers and percentages.

The GraphPad Prism computer program (version 5.0) was used for statistical analysis. The Mann–Whitney test was employed for nonparametric values and Student's *t*-test for parametric values, with the level of statistical significance for all tests being set for p-values <0.05.

Results

Tables 1 and 2 show the absolute and relative numbers (percentages) and other data of lymphocytes in the Study and Control Groups.