Possible Correlation Between ANA Positivity and Neoplasms in the Elderly: Preliminary Results

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Abstract

This is a case-control study in which elderly patients were compared regarding the presence or absence of neoplasms, dependent or not on ANA positivity. The control was composed of those with negative ANA and the group of cases was composed with those with positive ANA in low to moderate titers. The information was collected from a database of the immunology sector of the Blood Center of the Marília Medical School, state of São Paulo, Brazil. We evaluated 31 ANA positive patients (cases) and 36 ANA negative (controls). An OR = 1.93 (95% CI= 0.66 – 5.65) was found indicating a favorable trend for the association between ANA positivity and the presence of neoplasms, but without statistical significance. We believe more research is needed with larger samples in the study of this subject.

Keywords: Antinucleus Factor; Coarse Speckled (CS); Drugs

Introduction

ANA (Antinucleus Factor) is a widely used test for screening for autoimmune diseases. It indicates the presence or absence of autoantibodies against antigens of several cellular compartments: nuclear, nucleolar, cytoplasmic, mitotic apparatus and chromosomal plaque [1].

In the last two decades, the most widely used technique is indirect immunofluorescence with HEp-2 cells, a human tumor cell of an epithelial lineage derived from laryngeal carcinoma that allows the visualization of various cell structures and phases of the cell cycle. Before this technique, the information was given by the ANA-IFI (antinucleus-indirect immunofluorescence antibodies) resulted from the recognition of antigens present in the nucleus of the hepatocyte by autoantibodies present in the serum of patients [1].

Certain fluorescence patterns are more specific to autoimmune disease, while others occur frequently in healthy individuals or in patients with other non-autoimmune diseases. Coarse speckled (CS) nuclear pattern and homogeneous (H) staining nuclear pattern are often associated with autoimmune diseases; the fine dense speckled staining nuclear (PFD) and reticulate coarse speckled staining nuclear (PGR) patterns are predominantly associated with individuals without any evidence of autoimmunity and the pleomorphic nuclear pattern is related to neoplastic conditions [1].

The test is performed when there is clinical evidence of autoimmune disease, but there are situations which the ANA is

positive even without signs and symptoms of autoimmune disease. In these cases, the following hypotheses should be considered: autoantibodies associated with chronic inflammatory diseases, transient autoimmune disorder (infection, drugs, cancer), familial trait of autoimmunity, minimal manifestation of an autoimmunity spectrum, early manifestation of autoimmune disease [2].

In addition, healthy individuals may present positive ANA in a prevalence ratio of approximately 3-15% [3]. The production of these autoantibodies is also age-dependent, increasing to 10-37% in healthy people over the age of 65 [3].

The test is done from the collection of a small amount of blood wich is sent to a laboratory where a fluorescent dye is added to the sample that identifies all circulating antibodies. After the antibodies are labeled, the blood is mixed in a container with a culture of human cells (HEp-2 cells). If there are autoantibodies, these will attach to human cell structures forming immunofluorescence patterns, being more than 20 different patterns [4].

This process is repeated over several dilutions of the blood, until the fluorescence disappears. Positive results are those that remain glowing after 40 dilutions. These are usually done in the following order: 1:40, 1:80, 1:160, 1:320, 1:640 and 1:1280 [4]. Then, 1:40 and 1:80 are considered low titers, 1:160 and 1:320 are considered moderate, and dilutions greater or equal to 1:640 are considered high titers [1].

In view of these observations we elaborate the following question:

Would there be any correlation between ANA positivity in low to moderate titers and the presence of neoplasms in the elderly?

Therefore, the objective of this research was to evaluate the possible correlation between ANA positivity in titers from 1/40 to 1/320 with the presence of neoplastic diseases in patients aged 60 years or more, referred to the Laboratory of Immunology of the Blood Center of the Marília Medical School (FAMEMA) located in the city of Marília, state of São Paulo in Brazil..

Methods

This is a case-control study (retrospective), comparing the elderly with positive ANA with low to moderate titration with the elderly with ANA negative, evaluating the presence or absence of neoplasia.

Inclusion criteria

Patients aged 60 years or more referred to the laboratory of the blood center of FAMEMA for ANA test whose results have been: ANA negative (<1/40) or positive in titers from 1/40 to 1/320.

Exclusion criteria

ANA test with titers > 1/320; presence of autoimmune rheumatic diseases, infectious diseases, use of the following drugs: hydralazine, procainamide, isoniazid, methyldopa, chlorpromazine, quinidine, minocycline, family history of autoimmune diseases.

The study was developed through retrospective results, collected in a database of the immunology sector of the blood center of the Marília Medical School, of patients aged over 60 years, referred to the Laboratory of Immunology of the Blood Center of Marília-FAMEMA.

The sampling was by convenience: 36 ANA negative patients and 31 ANA positive patients at low to moderate titers, all aged 60 years or older:

- Group 1 (case) - Elderly patients ANA (+) with titrations from 1/40 to 1/320 with their respective patterns, verifying the presence or absence of neoplastic diseases.
- Group 2 (control) - Elderly patients ANA (-) evaluating the presence or absence of neoplastic diseases.

The results obtained were statistically analyzed as follows:

Quantitative data were presented as means and standard deviations and qualitative data (categorical variables) as absolute numbers and percentage of the total. The statistical calculation used for the possible associations was the odds ratio (OR) for a 95% confidence interval (95% CI). The study was approved by the Medical Ethics in Research Committee of FAMEMA and the study was conducted according to the recommendations of the Declaration of Helsinki.

Results

The general characteristics of the analyzed sample are presented in table 1 below.

Characteristic	ANA positive Nº(%)	ANA negative Nº(%)
Total	31	36
Men	12 (38.7%)	13 (36.1%)
Women	19 (61.2%)	23 (63.8%)
Age (years)	69.8 ± 7	68.8 ± 7.7
Neoplasias	11 (35.4%)	8 (22.2%)
Digestive system	1	0
Breast	2	1
Gynecological	1	1
Hematologic/Lymphoma	4	6
Head and neck	1	0
Skin	2	0

Table 1: General characteristics of sample.

We found the following association values between ANA positivity and the presence of neoplasia: OR = 1.93 (95% CI = 0.66-5.65).

Discussion and Conclusion

This study sought to explore a subject little researched wich has a growing interest due to the increase in the elderly population in our society as well as the known higher prevalence of both positive ANA in low titers and neoplasms in this age group. There was a favorable correlation between the ANA in low to moderate titers and the presence of neoplasms in the elderly, but without statistical significance, since the confidence interval (95%CI) of the odds ratio (OR) encompassed the value 1.

In general, patients with autoimmune diseases tend to have moderate and high titers, while healthy individuals and those with other non-autoimmune comorbidities with positive ANA tend to have lower titers. Therefore, ANA positivity in low to moderate titers can be seen in patients with neoplasms, age over 60 years, rheumatoid arthritis, progressive systemic sclerosis, discoid lupus, necrotizing vasculitis, Sjogren's syndrome, chronic active hepatitis, tuberculosis, and systemic lupus erythematosus due to the use of some types of medications. In systemic lupus erythematosus, ANA usually presents with titers greater than 1:160 [5].

In a study of 216 patients with a recent diagnosis of chronic lymphocytic leukemia (CLL), ANA positive was observed in 30

(13.8%) patients, much higher than that which was reported in healthy people (5.6%-8.5%) [6].

Thus, it is observed that neoplasms and ANA can influence each other. If on the one hand the immune system plays a defense role, preventing the emergence of neoplastic cells, on the other hand, mutagenic cancer cells have several antigenic proteins unknown to the immune system. Those that are not recognized as their own (autoantigens) can lead to the development of autoimmune phenomena (emergence of autoantibodies) [7].

According to the Brazilian Institute of Geography and Statistics (IBGE) the number of elderly has been growing, and in 2017, there was an increase of 18% of the elderly population in five years [8]. In addition, there is a high prevalence of neoplasms in the elderly, and 70% of all types of them will occur in individuals aged 65 years or more by the year 2023 [9].

Therefore, we believe that more studies and larger samples are needed which will allow more reliable conclusions on this subject of great interest to science.

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