

FREQUENCY OF ANTINUCLEAR AUTOANTIBODIES AMONG HEALTHY VOLUNTEERS

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ABSTRACT

Objective: To determine the frequency of antinuclear autoantibodies (ANA) among healthy individuals.

Study Design: Descriptive study.

Place and Duration of Study: Combined Military Hospital, Chunian Cantt and Armed Forces Institute of Pathology, Rawalpindi, from September to December 2009.

Subjects and Methods: Serum samples were collected from healthy volunteers (after informed consent) through non probability convenience sampling. Antinuclear antibodies were detected by indirect immunofluorescence. Titer of a positive sample was determined by serial dilution. Data was analyzed for frequency and percentage of positive samples.

Results: A total of 100 volunteers (50 males and 50 females) were inducted in study. Their age ranged from 2 years to 75 years. Out of these, 3 (3%) volunteers were found to be positive for ANA, though in low titers, i.e., less than 1:10.

Conclusion: Antinuclear autoantibodies are found in approximately 3% of healthy individuals. In the absence of symptoms, a low titer of ANA may not be of much significance as it may be found in healthy people as well. But in appropriate clinical settings, a positive ANA in high titer should be further investigated.

Keywords: Antinuclear autoantibodies, Connective tissue disorders, Systemic lupus erythematosus.

INTRODUCTION

Antinuclear autoantibodies (ANA) are commonly used for screening, diagnosis and monitoring of connective tissue disorders (CTD) such as systemic lupus erythematosus (SLE), progressive systemic sclerosis (PSS), Sjogren's syndrome (SS), polymyositis (PM) and mixed connective tissue disease (MCTD).¹ This is a group of autoantibodies directed against different specificities inside the nucleus, seen as different morphological patterns of ANA by indirect immunofluorescent staining². ANA constitute a part of American College of Rheumatology criteria for diagnosis of SLE^{3,4}. However, ANA have been found in healthy people, in patients with non-rheumatic conditions and in infections, though in lower titers than those in autoimmune diseases^{5,6}. Besides, in normal population, both the percentage of positive tests and the titer of ANA rise with age⁷. Various studies have

shown different frequency of these autoantibodies in healthy population.^{8,9} As both genetic and environmental factors can influence autoantibody production¹⁰, the frequency of ANA in healthy individuals may be different in our population. The present study was, thus, aimed to determine the frequency of antinuclear autoantibodies in healthy individuals.

SUBJECTS AND METHODS

This descriptive study was carried out at Combined Military Hospital, Chunian Cantt and Armed Forces Institute of Pathology, Rawalpindi, from September to December 2009. Serum samples were collected from 100 healthy volunteers (after informed consent) through non probability convenience sampling. All persons included in the study had their baseline investigations (blood complete picture, urine routine examination and chest x-ray) within normal range for their age and gender. Any person with a history of an illness during the past 3 months was excluded. Serum was separated and stored at -4°C until sent to reference laboratory (Armed Forces Institute of

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Pathology) for analysis. Antinuclear antibodies were detected (at a screening dilution of 1:10) by indirect immunofluorescence technique using HEp-2 cells as substrate, along with positive and negative controls. Titer of a positive sample was determined by serial dilution. Data was entered in SPSS version 15.0 and analyzed for frequency and percentage of positive samples.

Descriptive statistics were used to describe the data i.e mean and standard deviation (SD) for quantitative variables and frequency along with percentage for qualitative variables.

RESULTS

A total of 100 volunteers were inducted in this study and screened for antinuclear autoantibodies with equal gender distribution (50 males and 50 females). Their age ranged from 2 to 75 years (mean \pm SD 37 ± 14). Out of these, 3 (3%) volunteers were found to be positive for ANA (Fig). Two males were positive aged 65 and 38 years while one female positive was 52 years of age (Table). However, all samples were positive in low titer only (less than 1:10, after screening dilution). Titer over 1:10 was not detected among positive samples.

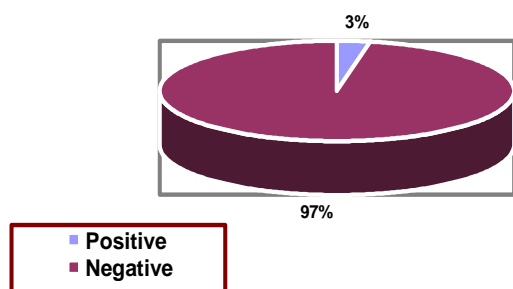


Figure: Frequency of ANA among healthy volunteers

Table: Frequency of ANA n=100

| | Males | Females | Total |
|----------|-------|---------|----------|
| Positive | 2 | 1 | 3 (3%) |
| Negative | 48 | 49 | 97 (97%) |

DISCUSSION

Antinuclear autoantibodies constitute a group of autoantibodies that represent a hallmark in diagnosis of a wide variety of autoimmune disorders particularly SLE, so that a negative ANA test makes the diagnosis of SLE highly unlikely.¹¹ It is useful in screening for

autoimmune disorders due to its high sensitivity. High titer ANA indicates a need to complement the investigation with tests for other autoantibodies such as anti dsDNA antibodies, anti Ro, anti La and anti Smith antibodies¹². Wide variety of methods are available for ANA detection like indirect immunofluorescence (IIF), enzyme linked immunosorbent assays (ELISA) and agglutination methods, each carrying its own merits and demerits¹³. Since ANA is amongst the most frequently advised immunological tests, it is important to know that in low titers, ANA can become positive in non-immunological conditions, infections, and even in healthy elderly individuals.^{5,6}

This study was undertaken to determine frequency of ANA among healthy individuals in our population. Among 100 subjects studied, ANA was positive among 3 (3%) individuals, though none had any manifestation of an autoimmune disorder. This constitutes a frequency of 3% in our general population. Various studies have revealed different ANA frequencies ranging from 4% to 22.6%.⁵ Ghosh et al have found ANA frequency of 4.3% among healthy individuals in India⁸ while ANA frequency in Saudi population was 4.2%.⁹ However, Fernandez et al have determined ANA frequency of 22.6% among healthy blood donors in Brazil.⁵ Thus our results are consistent with studies in our region indicating ANA frequency in healthy population to be around 3%. A positive ANA by itself is not sufficient to establish diagnosis of an autoimmune disorder¹⁴, rather a significant number of healthy population carry antinuclear autoantibodies in their blood without disease manifestations, albeit in low titer.^{5,8,9}

CONCLUSION

Antinuclear autoantibodies are found in approximately 3% of healthy individuals. Thus a low titer of ANA may not be of significant clinical value but in presence of appropriate clinical symptoms, a positive ANA should be further investigated with ANA titer and other laboratory tests (anti dsDNA antibodies, anti ENA antibodies, complement levels etc).

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