Clinical and Experimental Rheumatology 2004; 22: 227-229.

Antipolymer antibodies in Danish fibromyalgia patients

B. Jensen[1,2], I. Hechmann Wittrup[1], A. Wiik[3], H. Bliddal[1], S. Friis[2], J.K. McLaughlin[4,5], B. Danneskiold-Samsøe[1], J.H. Olsen[2,5]

[1]Parker Institute, Frederiksberg Hospital, Frederiksberg, Denmark; [2]Institute of Cancer Epidemiology, Danish Cancer Society, Copenhagen; [3]Department of Autoimmunology, Statens Serum Institut, Copenhagen, Denmark; [4]International Epidemiology Institute, Rockville, Maryland; [5]Department of Medicine, Vanderbilt University Medical Center, Vanderbilt-Ingram Cancer Center, Nashville, Tennessee, USA.

Bente Jensen, MD, PhD; Irene Hechmann Wittrup, MD, PhD; Allan Wiik, MD, DMSc; Henning Bliddal, MD, DMSc; Søren Friis, MD; Joseph K. McLaughlin, PhD; Bente Danneskiold-Samsøe, MD, DMSc; Jørgen H. Olsen, MD, DMSc.

Please address correspondence to: Bente Jensen, MD, PhD, P.G. Ramms Allé no. 1, 3 th., 2000 Frederiksberg, Copenhagen, Denmark. E-mail: bjensen@aab11.dk (reprints will not be available from the author)

Received on March 3, 2003; accepted in revised from on December 18, 2003.

Key words: Antipolymer antibodies, fibromyalgia, symptom severity, myalgic score, antinuclear antibodies

ABSTRACT

Objective. *To use a new immunologic assay to investigate antipolymer antibody (APA) levels in women with fibromyalgia (FM).*

Methods. *The study population consisted of 35 patients with FM and 129 controls. The controls were selected based on a prior history of breast surgery and the presence or absence of a prior hospital diagnosis of soft tissue rheumatism. Study subjects underwent blood sampling, including tests for antinuclear antibodies (ANA) and APA, a clinical examination, and an interview focusing on rheumatic complaints and self-reported disability. The severity of rheumatic symptoms/signs was scored from 1 (= none) to 5 (= severe) based on the clinical examination and the interview.*

Results. *FM patients in this study represented a broad spectrum of disease severity, with the majority having mild symptoms. FM patients had a higher symptom severity and myalgic scores than controls ($p < 0.001$ for both variables). Adjusting for symptom severity, a weak positive association between APA levels and FM was observed ($p = 0.08$). The APA level was inversely associated with age, i.e., decreasing APA levels were seen with increasing age ($p = 0.008$).*

Conclusion. *FM patients tended to have slightly higher APA levels than controls when adjusted for symptom severity. APA levels declined with age, a finding that has not been reported previously. The APA test and its clinical relevance should be evaluated in future studies.*

Introduction

Fibromyalgia (FM) is a non-articular rheumatic syndrome characterized by chronic, widespread pain and marked tenderness to pressure at specific sites on physical examination (1). Except for the presence of tender points, physical findings are frequently absent, and there is no known laboratory test that is diagnostic for FM. Although there are validated health status instruments that measure the impact of FM, standardized outcomes to define the severity of FM have not been established (2).

The pathogenic mechanisms underlying FM are poorly understood. Some investigators have argued for a relation to inflammatory rheumatic diseases, based upon the presence of antinuclear antibodies (ANA) and symptoms indicative of connective tissue disease among FM patients (3). However, current evidence does not support an inflammatory rheumatic nature of FM (4, 5).

Recently an antipolymer antibody (APA) assay has been developed (6). Antibodies to the partially polymerized acrylamide antigen used in this assay were found in the sera of symptomatic women with silicone breast implants (SBIs). The level of APA was reported to be associated with the severity of clinical symptoms in SBI recipients, and the test was proposed as an objective diagnostic tool for SBI-related complaints. The presence of APA has also been reported in FM patients without SBIs, with the highest APA levels observed in patients with the most severe symptoms (7). Many of the symptoms reported by women with SBIs appear to be similar if not identical to those observed in patients with FM (8).

The objectives of this study were to compare APA levels as measured by this new assay in women with FM and control subjects and to examine whether APA levels are associated with the severity of rheumatic symptoms in women without SBIs.

Materials and methods

The study population was identified through the Danish National Hospital Register of Patients (NRP), the Central Population Register (CPR) and from the fibromyalgia files of the Department of Rheumatology, Frederiksberg Hospital. Study subjects were 35 women with a diagnosis of FM who had not undergone any type of breast surgery, and four subgroups of control women without FM selected according to a history of breast surgery (breast reduction/no breast surgery) and the presence or absence of a prior hospital diagnosis of non-specific soft tissue muscular rheumatism (groups A, B, C, and D; total n = 129) (Table I). Validation of the diagnosis of muscular rheumatism

in two recent studies (9,10) revealed that the diagnosis was primarily based on simple localized soft tissue complaints.

The study participants underwent a thorough clinical examination, blood tests and an interview focusing on possible rheumatic complaints. Tender points located according to the American College of Rheumatology criteria (1) were noted. The following scoring system for grading the severity of pain upon pressure was used: 0, no pain; 1, mild pain without grimace; 2, spontaneous verbal reactions to pain and grimace; and 3, severe pain with withdrawal. A myalgic score was defined as the pain severity score multiplied by the respective number of tender points. The myalgic score could range from zero to 54. A Likert rheumatic scale for symptom severity ranging from 1 (no symptoms or signs) to 5 (severe symptoms and severe signs) was applied on the basis of self-reported symptoms and clinical signs. Standard blood tests indicative of inflammatory rheumatic diseases and the APA test were performed on all participants. The APA kits and all chemical needed were provided by Dr. R.B. Wilson, Autoimmune Technologies, LLC (New Orleans, LA, USA). Optical density (OD) values were recorded on triplicates of each of the reference and patient samples. For details about the APA test we refer the reader to the study by Wilson *et al*. (7).

Statistical methods

Comparisons between FM cases and controls were analyzed by the Mann-Whitney test and the Chi-Square test. Correlation analyses were performed by Spearman's rank correlation. APA measurements were calculated as the average of the log-transformed values of OD minus the average of the log-transformed kit-specific blank values. The analyses of variables possibly related to APA measurements were performed by analysis of variance (ANOVA) and analysis of covariance (ANCOVA). We evaluated whether the sampling procedure (controls selected according to history of breast surgery (breast reduction/no breast surgery) and the presence or absence of a prior diagnosis of muscular rheumatism) influenced the results. The controls were classified by the combination of two categorical (yes/no) variables, breast reduction and muscular rheumatism. An analysis of variance that included these two variables as well as the interaction between them was performed.

Successive exclusion of these factors showed no association with the APA measurements (all $p > 0.20$), which allowed us to combine the four subgroups of controls into a single control group for subsequent analyses. In the covariance analysis we considered the categorical variables: i) FM, ii) symptom severity score, and iii) ANA; and the linear variables iv) age and v) myalgic score as covariates. All analyses included the kit number (1-9) as a categorical variable to adjust for between-kit variation. The statistical analyses were performed using the SPSS statistical package, version 9.

Results

Characteristics of FM cases as well as the four control subgroups (groups A, B, C and D) are presented in Table I. Fifty-six percent of the FM patients had mild (symptom severity score 3) symptoms and 32% and 12% had symptom severity scores of 4 (moderate) and 5 (severe) (data not shown). Significant differences in the symptom severity scores and myalgic scores were observed between FM patients and controls ($p < 0.001$ for both variables) (Table I). In the crude analysis (Mann-Whitney test) APA levels were similar among FM patients and controls (Table I). When adjusting for the symptom severity score, myalgic score, age, ANA, and kit number women with FM had slightly a higher APA level than controls ($p = 0.08$) (Table I). Age was significantly associated with APA levels ($p = 0.008$). A scatter plot with the correlation between age and APA measurements showed that APA levels declined with age (Fig. 1).

ANA positivity was similar in the two groups (FM: 11%, control group: 13%) ($p = 0.73$) (data not shown). All selected women with a diagnosis of FM

Table I. Characteristics of the study groups.

	Fibromyalgia (FM)	A. Breast reduction (+r)	B. Breast reduction (-r)	C. No breast surgery (+r)	D. No breast surgery (-r)	Combined control group (A+B+C+D)	FM vs combined control group	
	N=35	N=24	N=26	N=23	N=56	N=129	P*	P**
Age in yrs, median (range)	45 (30-74)	43 (30-70)	45 (33-72)	43 (34-61)	48 (33-74)	45 (30-74)	0.52	-
Likert symptom severity score, median (range)	3 (3-5)	3 (1-5)	2 (1-3)	3 (2-4)	2 (1-4)	2 (1-5)	<0.001	-
Myalgic score, mean (SD)	34.6 (4.3)	22 (11.8)	9.6 (10.2)	19 (9.9)	7.3 (7.4)	12.6 (11.1)	<0.001	-
APAmean (SD) (background subtracted) [1]	0.4690 (0.3637)	0.3576 (0.2300)	0.5717 (0.4540)	0.5263 (0.3387)	0.3311 (0.2368)	0.4193 (0.3232)	0.84	0.08

APA: antipolymer antibodies.
(+r) and (-r) indicate women with and without a prior diagnosis of muscular rheumatism
*p: Mann-Whitney test used for comparisons; **p: Analysis of covariance with adjustment for symptom severity score, myalgic score, ANA, age, and kit number
[1]APAvalues based on analyses with subtraction of background values of APA

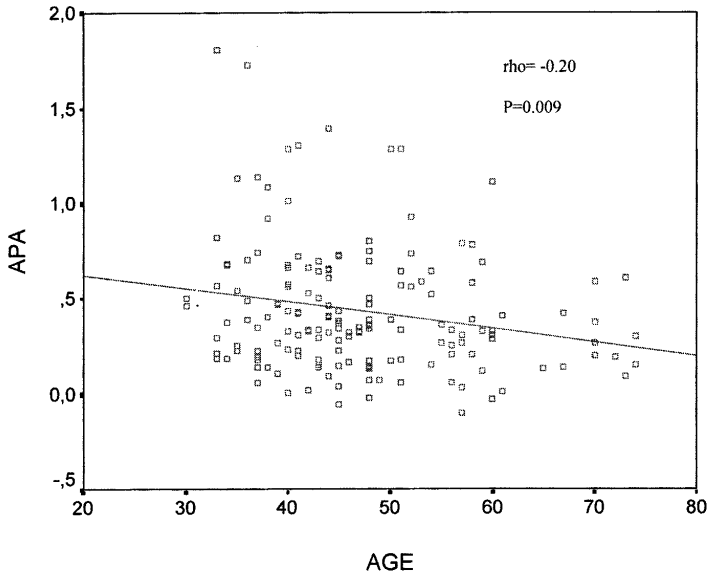

Fig. 1. Correlation between antipolymer antibodies (APA) and age (Spearman's correlation coefficient). The APA values are mean values of log-transformed APA values (the mean of the log-transformed blank values were subtracted).

retained their diagnosis at the clinical examination, while none of the control subjects fulfilled the criteria for FM. One woman with FM and 14 controls had inflammatory rheumatic diseases (including rheumatoid arthritis, primary systemic sclerosis, systemic lupus eythematosus, spondylarthropathies, polyarteritis nodosa, and gout; $p = 0.26$). In all measurements, we observed a substantial, albeit not statistically significant, between kit variation.

Discussion

FM is a syndrome characterized by widespread pain and disability (1,2). The observation of higher myalgic and symptom severity scores among FM patients compared with controls was thus to be expected. When adjusting for symptom severity, a slightly higher APA level was observed among FM patients compared with controls. This result is parallel to the finding in a recent study by Wilson *et al*. who reported higher APA values among FM patients with severe FM than among those with mild symptoms (7). In our study the majority of FM patients had mild symptoms, thus limiting our ability to examine APA level by symptom severity. Our study is not directly comparable to that of Wilson *et al*. (7), who used a cutoff to define APA positivity and defined APA based on both FM patients and subjects with CTDs.

Our control group was composed of both healthy women and women with rheumatic diseases. Furthermore, some of them had undergone breast reduction surgery or had previously been hospitalized for soft tissue rheumatism. There was a higher frequency of inflammatory rheumatic diseases among controls compared with FM patients; however, when we excluded women with these conditions (one from the FM group and 14 from the control group), APA levels were similar in the two groups.

We found that the APA level was not related to the myalgic score or to ANA positivity. The last finding is in line with Tenenbaum *et al.* (6). FM patients did not have a higher prevalence of ANA positivity compared with controls.

In general, the level of several serum autoantibodies, e.g. ANA, increases abnormally with age (11). Our finding of a decrease in APA level with increasing age may indicate that APA does not reflect an autoantibody directed against an unrecognized self-antigen. This indicates that a potential link to autoimmune disorders is less likely. However, the clinical implications of this finding remain to be clarified.

In conclusion, we found a weak positive association between FM and APA level when controlled for the symptom severity score. Because of the small proportion of FM patients with severe symptoms in our study, and the substantial between-kit variation in the APA measurements, we suggest that the APA test should be validated further. In addition, studies seeking to characterize the antigen eliciting the APA response appear warranted. Clinical outcomes associated with the presence of APA remain to be established. The finding of decreasing APA levels with age warrants further exploration. Finally, standardized methods to evaluate the severity of FM symptoms should be developed.

References

1. WOLFE F, SMYTHE HA, YUNUS MB *et al.*: The American College of Rheumatology 1990. Criteria for the classification of fibromyalgia. Report of the Multicenter Criteria Committee. *Arthritis Rheum* 1990; 33: 160-72.
2. WOLFE F, AARFLOT T, BRUUSGAARD D *et al.*: Fibromyalgia and disability. *Scand J Rheumatol* 1995; 24: 112-8.
3. DINERMAN H, GOLDENBERG DL, FELSON T: A prospective evaluation of 118 patients with the fibromyalgia syndrome: Prevalence of Raynaud's phenomenon, sicca symptoms, ANA, low complement, and Ig deposition at the dermo-epidermal junction. *J Rheumatol* 1986; 13: 368-73.
4. BENGTSSON A, ERNERUDH J, VRETHEM M, SKOGH T: Absence of autoantibodies in primary fibromyalgia. *J Rheumatol* 1990; 17: 1682-3.
5. KOGSTAD O: Primary fibromyalgia syndrome. Subgroups of inflammatory rheumatic nature? *Scand J Rheumatol* 1988; 17: 154.
6. TENENBAUM SA, RICE JC, ESPINOZA LR, CUÉLLAR ML, PLYMALE DR, SANDER DM: Use of antipolymer antibody assay in recipients of silicone breast implants. *Lancet* 1997; 349: 449-54.
7. WILSON RB, GLUCK OS, TESSER JRP, RICE JC, MEYER A, BRIDGES AJ: Antipolymer antibody reactivity in a subset of patients with fibromyalgia correlates with severity. *J Rheumatol* 1999; 26: 402-7.
8. WOLFE F: "Silicone related symptoms" are common in patients with fibromyalgia: no evidence for a new disease. *J Rheumatol* 1999; 26: 1172-5.
9. JENSEN B, KJØLLER K, McLAUGHLIN JK *et al.*: Muscular rheumatism following breast surgery in Denmark. *Clin Exp Rheumatol* 2001; 19: 229.
10. JENSEN B, BLIDDAL H, WITTRUP IH *et al.*: Rheumatic manifestations in Danish women with silicone breast implants. *Clin Rheumatol* 2001; 20: 345-52.
11. SLATER CA, DAVIS RB, SHMERLING RH: Antinuclear antibody testing. A study of clinical utility. *Arch Int Med* 1996; 156: 1421-5.