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SEROLOGICAL PROFILE (RHEUMATOID FACTOR AND ANTI-CITRULLINATED PEPTIDES ANTIBODIES) IN A GROUP OF ALGERIAN PATIENTS WITH RHEUMATOID ARTHRITIS

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Abstract

Introduction: Rheumatoid arthritis (RA) is the most common inflammatory rheumatism. It is a multifactorial autoimmune disease characterized serologically by the presence of auto-antibodies: rheumatoid factor (RF) and anti-citrullinated peptides antibodies (ACPA) .The objective of this study is to establish a serological profile in Algerian RA patients and correlate it to disease activity. **Patients and Methods:** Our case-control study covered 343 patients divided into 49 male and 294 female. Auto-antibodies was carried out using the laser nephelometry technique for the RF and by the quantitative enzyme immunoassay technique ELISA 3rd generation test for the ACPAs. **Results:** Our results showed that among 314 patients, 67.8% were positive for the RF, with an average serum titer of 231.4 ± 506.5 IU /mL. The ACPA search returned positive for 79.3% of patients, with an average serum concentration of 217 ± 337.5 IU / mL. Our analysis showed that out of a total of 315 RA, 269 patients or 85.39% are positive (RF positive (+) and / or ACPA +), while 46 patients, or 14.60%, are negative (RF and ACPA negative). **Conclusion:** Our results showed a significant sensitivity of ACPA compared to RF. ACPAs have better diagnostic value than RF. Benefit of the RF/ACPA association to increase specificity. Autoantibody levels are associated with the severity of the disease.

Keywords: Rheumatoid Arthritis; Rheumatoid Factor; Anti-citrullinated Peptides Antibodies; DAS28 (Disease Activity Score)

I. INTRODUCTION

Rheumatoid arthritis (RA) is the most common chronic inflammatory rheumatism in adults. It is a multifactorial autoimmune disease, where environmental and hormonal factors contribute to the breakdown of tolerance in genetically predisposed subjects [1].

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It affects bilaterally and usually symmetrically the peripheral joints of the limbs and is characterized in particular by the pseudo-tumoral proliferation of the synovial tissue (rheumatoid pannus) and rapid progressive destruction of the articular structures. [2].

In Algeria, estimating the prevalence of RA is still uncertain and difficult, due to the lack of a register or sufficiently exhaustive medico-administrative databases. On a 2010 prospective study conducted in Barika (municipality of the wilaya of Batna), populated by 125,253 subjects with 52,504 adults (26,358 men and 26,146 women). The prevalence of RA locally reported 0.13% and 0.15% by extrapolation to the Algerian population [3].

RA is characterized by the activation of self-reactive T and B clones and serologically by the presence of auto-antibodies: rheumatoid factor (RF) and anti-citrullinated peptide antibodies or anti-citrullinated peptide antibodies (ACPA) both introduced as diagnostic criteria by the American College of Rheumatology (ACR) in 1987 and the European League Against Rheumatism (EULAR) in 2010 respectively [4].

The association RF and ACPA is highly recommended to increase the diagnostic relevance. ACPAs are very specific (>98%) and very sensitive (80%). In addition to their predictive value, they constitute an index of severe progression of the disease (prognostic markers) [5].

II. MATERIALS

Table 1: Demographic Characteristics of the RA Population

characteristics	All patients N=343	women N=294	men N=49
Age (years)	48,4 ± 13	47,7 ± 13	52,3 ± 12,7
Sex ratio (M/W)	1/6	/	/
Disease duration (years) (N=266)	12,47 ± 8,41	12,77 ± 8,38	10,56 ± 8,49
Old RA (>2 Years)	245/266 (92,10%)	216/230 (93,9%)	29/36 (80,6%)
Early RA (≤ 2 Years)	21/266 (7, 89%)	14/230 (6, 1%)	7/36 (19,4%)

Patients and Healthy Subjects (Controls)

This is a case-control study conducted in our laboratory on 688 subjects of both sexes, unrelated and aged over 15 years. Including 343 patients with rheumatoid arthritis (RA) and 345 healthy subjects

The 343 patients were recruited by two rheumatology departments (that of the university hospital center (UHC) Béni-Messous and that of the specialized hospital establishment (EHS) of Ben Aknoun). The diagnosis of RA was retained according to the ACR 1987 criteria (the patient must meet at least 4 of the 7 criteria). We have, for each patient, a detailed clinical and biological sheet listing all the inclusion criteria. They are divided into 49 male subjects and 294 female subjects (Table 1).

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345 healthy subjects, including 282 female subjects and 63 male subjects, were included in our study (Table 2). They were recruited from the blood transfusion center and the central laboratory of medical biology of the CHU Béni-Messous

These subjects are unrelated and free of any rheumatic or other inflammatory pathologies, with no personal or family history of autoimmunity. They were recruited on the basis of negative inflammatory and autoimmunity assessments

Table 2: Demographic Characteristics of the Control Population

characteristics	healthy subjects (controls) N=345
Age (years)	37 ± 11,6
Women N (%)	282 (81,7%)
Men N (%)	63 (18,3%)
Sex ratio (M/W)	1/5

III. METHODS

III-1. Clinical study:

III-1.1. Assessment of Clinical Activity

The DAS28 (Disease Activity Score) is a composite index of RA activity developed by EULAR. The calculation of the DAS 28-CRP of patients with RA was determined by rheumatologists in 271 patients. RA is low activity if the activity level is ≤ 3.2, moderately active if 3.2 < DAS 28 ≤ 5.1 and very active if > 5.1.

III- 1.2. Assessment of physical functional capacity (Health Assessment Questionnaire) (HAQ):

The calculation of the HAQ index was performed by rheumatologists in 143 RA patients. The overall HAQ value is calculated based on the average of the twenty questions. The index can integrate values between 0 and 3: 0 = no limitation of physical capacities; and 3 = extreme limitation.

III-2. Inflammatory biological assessment:

III-2.1. Determination of sedimentation rate (ESR)

III-2.2. Assay of C- reactive protein (CRP): Carried out by the laser nephelemetry technique, using the BN ProSpec automaton from the manufacturer Siemens.

III-3. Autoimmunity assessment:

III-3.1. Assay of rheumatoid factor (RF): carried out by laser nephelemetry: using the BN ProSpec automaton from the manufacturer Siemens.

III-3.2. Dosage of 3rd generation anti-cyclic citrullinated peptide antibodies INOVA (Quanta Lite CCP3 IgG): performs Quantitative immunoenzymatic assay.

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IV. RESULTS

IV -1: General Characteristics of the Population Studied

Our cohort of patients with RA is characterized by a female predominance of 85.71%, with an average age of 48.4 ±13 years and with a sex ratio of 1/6. As for the control population, the average age is 37 ± 11.6 years, a sex ratio of 1/5 and 81.7% of the subjects are women (Table 3).The duration of the disease in our patients is 12.47 ± 8.41 years, mostly old RA 92.10% (greater than 2 years). Biologically, we find an inflammatory syndrome in our patients, with a sedimentation rate of 44.1 ± 29.3 mm the first hour (average) and an average CRP of 5.8 ± 14.2 mg/L (Table 3).

The disease activity assessed by the DAS28-CRP score is 4.51 ± 1.41(average) with a mean functional disability of 1.33 ± 0.83 (Table 3).

Table 3: General Characteristics of RA Patients and Control Subjects

characteristics	RA Patients N=343	Controls N=345
Age (Years)	**48,4** ± 13	37 ± 11,6
Sex ratio (M/W)	**1/6**	1/5
Women	294 (85,71%)	282 (81,7%)
Disease duration (Years)(N=266)	**12,47** ± 8,41	/
Early RA (≤Years)	21/266 (7,89%)	**/**
Old RA (> 2 Years)	245/266 **(92,10%)**	**/**
ESR (mm/h) (N=308)	44,1 ± 29,3	9,53 ± 11,58
CRP (mg/L) (N=319)	5,8 ± 14,2	/
DAS28-CRP (N=271)	**4,51** ± 1,41	/
HAQ (N=143)	1,33 ± 0,83	/

IV-1.a. Distribution of patients according to disease activity

RA activity was assessed by calculating the DAS28 index. It was estimated for 271 subjects. The majority of them, 46.86%, had moderately active disease. 33.94% of cases had very active RA; while more than 19% of patients had weakly active RA. These results are shown in Figure 1.

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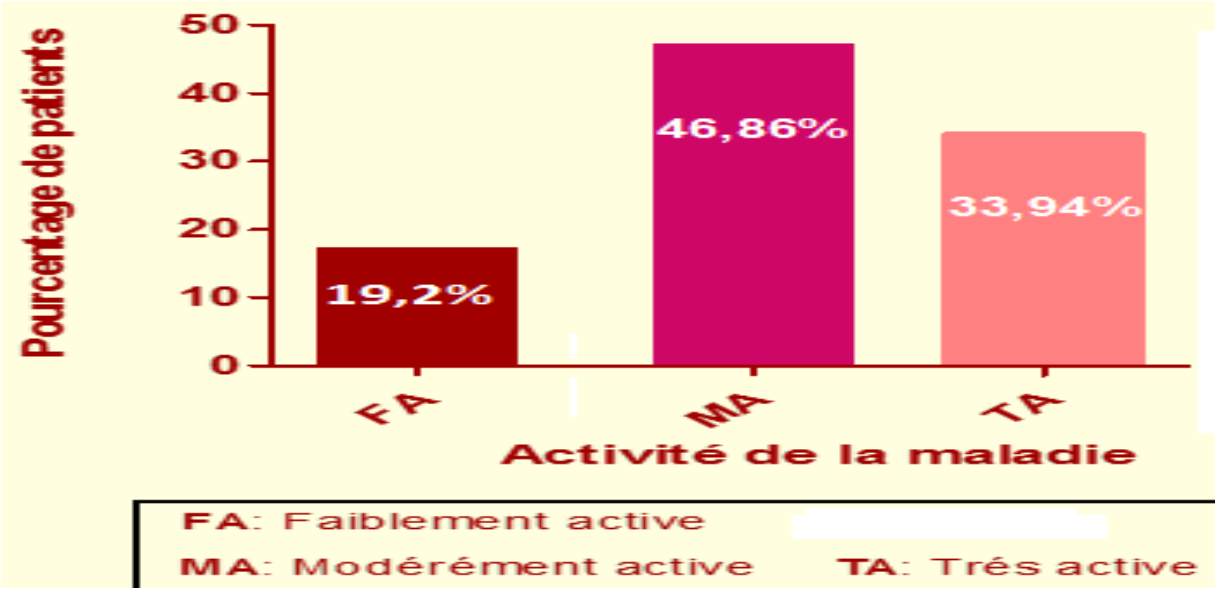

Figure 1: Distribution of Patients According to the DAS28 Score

IV-1.b. Functional Capacity According to Disease Activity

The functional capacity of RA patients was assessed by the HAQ score in 143 patients. Our results showed that it is significantly associated with disease activity in our cohort (p = 0.005) (Figure. 2).

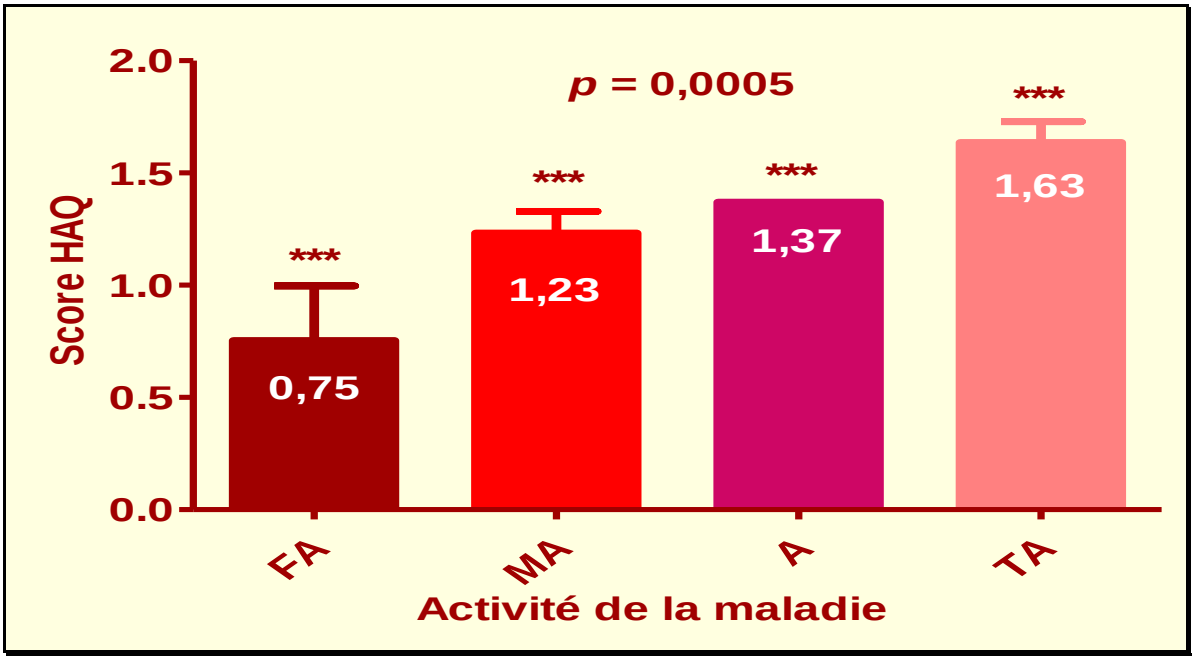

Figure 2: HAQ Score According to Disease Activity

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IV-1.c. The Average Age of RA Patients by Gender

The average age of rheumatoid arthritis in our cohort is 48.4 ± 13 years. Our results showed that the average age (between the two sexes) is statistically different; it is 52.3 ± 12.7 for men and 47.7 ± 13 for women (Figure 3).

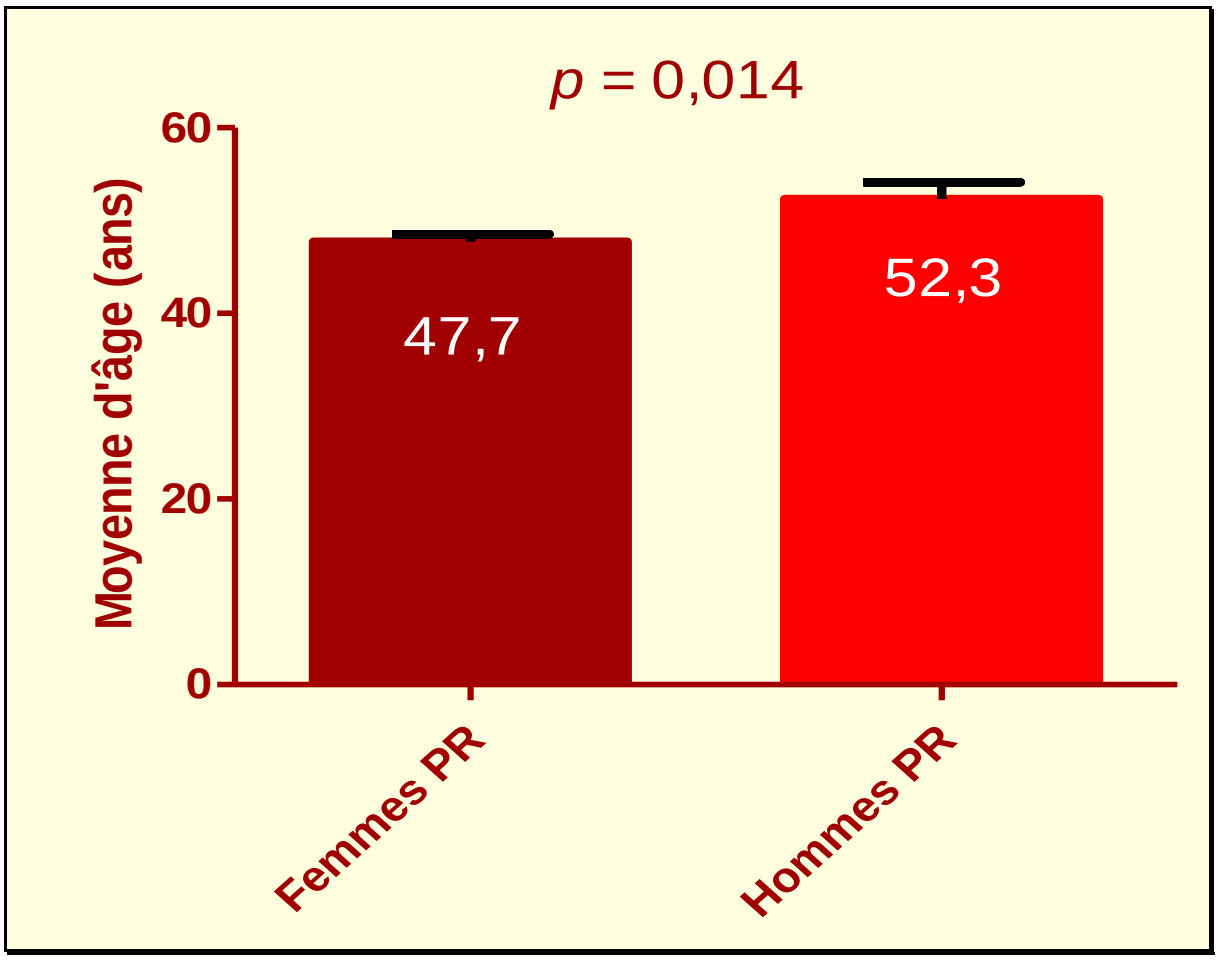

Figure 3: Average Age of RA Patients by Gender

IV-2: Serological characteristics of the population studied

Our results showed that among 314 patients, 67.8% are positive for total RF, with a mean serum titer of 231.4 ± 506.5 IU/mL. (Table 4).

The search for ACPA came back positive for 79.3% of the patients, with an average serum concentration of 217 ± 337.5 IU/mL (Table 4). Our analysis showed that out of a total of 315 PR, 269 patients or 85.39% are seropositive (RF positive (+) and/or ACPA +), while 46 patients, or 14.60%, are seronegative (RF and ACPA negative)

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Table 4: Serological Characteristics of Patients with RA.

Caractéristiques	RA Patients N = 343	Women N = 294	Men N = 49	*p*
Total RF positivity (≥ 20 UI/mL)	213/314 **(67,8%)**	182/270 (67,4%)	31/44 (70,5%)	0,688
RA Total Titers (UI/mL)	231,4 ± 506,5	225,8 ± 519,6	272,3 ± 404,7	0,21
ACPA positivity (N=329) (≥ 20 UI/mL)	261/329 **(79,3%)**	218/282 (77,3%)	43/47 (91,5%)	**0,026***
Titres ACPA (UI/mL)	**217** ± 337,5	202 ± 285,3	302,7 ± 553,1	0,11

V. DISCUSSION

Our cohort is characterised by a clear female predominance, with an M/F sex ratio of 1/6 and an average age of 48 years. These data are consistent with those found in the literature [6].

As for our study, the presence of RF has been correlated with a severe progression of RA in numerous studies [7].

Protein citrullination occurs during inflammation, keratinization and apoptosis. Its harmful consequence is the production of autoantibodies targeting citrullinated proteins which constitute epitropes for ACPAs, the production of antibodies against citrulline residues is quite specific for RA.

ACPAs are very interesting predictive markers of the severity of RA. Different teams have shown that among the markers that predict the severity of RA, ACPAs have the best odds ratio, which confirms the predictive value of these autoantibodies in structural damage and its aggravation by showing that this value is independent of other predictors [8].

Studies have established significant correlations between the presence and/or titer of ACPAs and various clinical, radiological or biological criteria of activity and/or severity of RA (rheumatoid nodules, CRP, circulating immune complexes, destruction joints…) and bone erosion after several years of development.

The detection of ACPA is therefore of major clinical interest, both because of their diagnostic value, from the early stages of RA, but also because of their prognostic value, making it possible to establish therapeutic strategies adapted to patients [8].

Our results showed a significant sensitivity of ACPA compared to RF. ACPA have better diagnostic value than RF in terms of sensitivity and specificity, as also demonstrated by Schattner's team [9].

Indeed, 79.3% of RA patients had a significantly positive rate of ACPA, against 67.8% of RA with positive RF. Subjects with high serology have a higher DAS28 index, same findings obtained in a Turkish study [10].

The simultaneous analysis of RF and ACPA shows that the combination of these two markers improves the specificity to reach 98.92%, hence the interest of assaying these

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two autoantibodies in the face of any suspicion of RA, these remarks are also supported by several studies. Including a meta-analysis [11, 12].

Furthermore, note that 14.6% of our patients are seronegative RA (negative ACPA and RF), despite the fact that our sick population consists essentially of old and proven RA, these are the true seronegative RA [13].

CRP is one of the parameters used in the calculation of the DAS 28 index, which makes it possible to assess the activity of the disease. This protein remains a marker of choice to know the inflammatory state of the patient, a correlation of the serological results with the index of evolution of the disease, (DAS28), was carried out. The results obtained demonstrated a clear positive correlation. Indeed, subjects with severe serology (raised serum autoantibody levels) had a higher DAS28 index. These facts are found in a study carried out by Solomon's team, which shows a significant correlation between the DAS 28 index and joint destruction [14], hence the interest in measuring this index in order to assess the severity and prognosis of the disease for better patient follow-up and optimization of treatment.

VI. DECLARATION OF INTERESTS

The authors declare no conflict of interest

VII. Conclusion

In summary, this study suggests that ACPA is more sensitive compared to RF. ACPAs have better diagnostic value than RF. Benefit of the RF/ACPA association to increase specificity. Autoantibody levels are associated with the severity of the disease.

Références

1) C Loreto, M Cross, B Williams, M Lassere, L March. Rheumatoid Arthritis. Best Practice & Research Clinical Rheumatology. 2010. 24 (6): 733‑45.

2) P Emery, B. McInnes, R. van Vollenhoven, M. C. Kraan. Clinical Identification and Treatment of a Rapidly Progressing Disease State in Patients with Rheumatoid Arthritis. Rheumatology . 2008. 47 (4): 392‑98

3) S Slimani, A Ladjouze et al. Polyarthrite rhumatoïde masculine et féminine : quelles différences ? Données à partir d'une étude multicentrique, 24ème Congrès de la SFR Décembre 2011.

4) D Aletaha, T Neogi, AJ. Silman, J Funovits, D T. Felson, O Clifton. Rheumatoid Arthritis Classification Criteria. Arthritis and Rheumatism. 2010. 62 (9): 2569‑81.

5) Y.W. Song et E.H. Kang. Autoantibodies in Rheumatoid Arthritis: Rheumatoid Factors and Anticitrullinated Protein Antibodies. QJM, 2010. 103 (3): 139‑46.

6) R Voskuhl. Sex differences in autoimmune diseases. Biology of Sex Differences. 2011, 2 (janv): 1.

7) B Combe et al. Facteurs prédictifs de la sévérité de la polyarthrite rhumatoïde. An Med Int. 2008. Vol 152 - N° 8 P. 522-526

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E-Publication: Online Open Access
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8) P Suwannalai, K Britsemmer, R Knevel, H Scherer, E W Nivine, H Levarht. Low-avidity anticitrullinated protein antibodies (ACPA) are associated with a higher rate of joint destruction in rheumatoid arthritis. Annals of the rheumatic diseases. 2012. 73 (1), 270-276

9) A Schattner. Anti-Cyclic Citrullinated Peptide Antibody Is a More Specific Test for Rheumatoid Arthritis than Rheumatoid Factor. *Evidence-Based Medicine.* 2007.12 (6): 183.

10) M Dedeoglu, U Gafuroglu, O Yilmaz and H Bodur.The relationship between hand grip and pinch strengths and disease activity, articular damage, pain, and disability in patients with rheumatoid arthritis. Turkish Journal of Rheumatology. 2013 June, Vol. 28, Issue 2, p. 69.

11) K Nishimura, D Sugiyama, Y Kogata, G Tsuji, T Nakazawa, S Kawano. Meta-analysis: diagnostic accuracy of anti-cyclic citrullinated peptide antibody and rheumatoid factor for rheumatoid arthritis. Ann Intern Med. 2007 Jun 5;146(11):797-808.

12) P Legoux. Polyarthrite rhumatoïde : les éléments biologiques, diagnostiques et pronostiques. Réalités en rhum. 2013 Juin; N 54 : 3-9**.**

13) TC Sang, HL Kwang. Clinical management of seronegative and seropositive rheumatoid arthritis: A comparative study. PLoS One. 2018; 13(4): e0195550.

14) DH Solomon, J Finkelstein, N Shadick, M LeBoff, C Winalski, M Stedman.The relationship between focal erosions and generalized osteoporosis in postmenopausal women with rheumatoid arthritis. Arthritis & rheumatism. Vol. 60, No. 6, 2009 June. p 1624–1631.