Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 4, 2021, Pages. 6756 – 6767
Received 05 March 2021; Accepted 01 April 2021.

Rheumatoid Arthritis and the Relationship of the Disease to Sex and Age and Some Hematological Variables for Affected Patients

Abeer Talib Abdulqader

Biology /Physiology ,Department of medical laboratory Techniques, Northern technical university, Al-dour technical institute, abeer.ta@ntu.edu.iq

Wijdan I.A. Abd-alwahab

University of Samarra, College of Education, Department of Biology, wijdan80@uosamarra.edu.iq

Bushra Ismail Ibrahim

Department of Biology, College of Education, University of Tikrit ,General Directorate of Education Salah Al-din, bushraismail856@gmail.com

Abstract

Rheumatoid Arthritis (RA) disease is a chronic systemic inflammatory disease and the reasons for its occurrence are not completely clear, and the disease targets the peripheral joints in an inconsistent manner, and its symptoms include the patient's feeling of fatigue, malaise, and stiffness in the morning in general, and its effect extends to sites located outside the joints Including the skin, the heart, the lungs and the eyes. And it causes joint damage in addition to side effects that lead to a disease that may sometimes cause death (Aaran *et al*, 2003). It is believed that the cause of the disease is infectious pathogens such as Mycoplasma, Epstein-Barr virus, and Parvovirus. However, none of the organisms has been confirmed as the cause of this disease. There are several autoimmune responses that have correlation with the disease, and these responses may be the first or second cause of the disease (Burtis and Ashwood, 1994). While other studies confirm that (RA) has an important genetic cause has been identified in 95% of patients. The occurrence of cell synovial overgrowth and endothelial cell activity during the onset of the disease lead to progression of uncontrolled infections and destruction or damage to cartilage and bones at the joint site. Genetic factors and a deviation or imbalance in the immune system contribute to the occurrence and development of the disease (Noskov *et al*, 2004). The incidence of the disease is about (3) cases per 10,000 people, and it affects all groups without exception and is more prevalent in the Native American population (American Indians), where the infection rate reaches between 5-6% and is less prevalent in the population of people of African descent (blacks). The disease may cause a shortening of the patient's life by (5-10) years than his peers, and it affects females by (2-3) times that of males, and his age period is an upper limit recorded

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 4, 2021, Pages. 6756 – 6767
Received 05 March 2021; Accepted 01 April 2021.

from (35-50) years, but this does not prevent Incidence in children and the elderly (Majithia and Geraci., 2007; Bessiere *et al*., 2001).

Introduction

Rheumatoid arthritis is a type of arthritis and it is one of the common diseases in contemporary and modern medicine, and the presence and distribution of lesions in ancient skeletons indicates that it occurred in North America 3000 years ago (Klippel *et al*, 2001).

The discovery of the rheumatoid factor, which is an autoantibody directed towards the active part of the immune globin IgG, in serum and synovial fluid began with a hypothesis made by Frank Billing in 1912, in Chicago, where he assumed that rheumatoid arthritis is a response to many chronic localized infections depending on the studies that followed this assumption Russel and his group in 1927 concluded, The rheumatoid arthritis results from infection with a special strain of streptococcus, and the researcher Martin in 1932 was not able to prove the role of bacteria in this case, but he explained that the rheumatoid serum has the ability to accompany suspensions of different types of bacteria, and Erik waaler in 1940 showed the ability of rheumatoid serum to analyze pellets Red blood of sheep using the Complement fixation test. Rose in 1947 demonstrated the ability of this serum to balance sheep's erythrocytes at high concentrations, the use of activated red blood cell agglutination assay as a diagnostic test, the use of the human globulin-covered latex clumping reaction in the diagnosis, and rheumatoid arthritis included in the group of associative tissue disease or collagen disease (Frank *et al*., 1995), In the late 1950's, molecular studies confirmed that the rheumatoid factor represents antibody molecules, and it was later observed that it specializes in determinants on its IgG immunoglobulin molecule.

Rheumatoid arthritis causes many changes and abnormalities, including damage to small vascular synovial cells represented by blockage of the vascular lumen, swelling of endothelial cells, moderate hyperplasia of the surface layer of endothelial cells, formation of gaps between these cells, congestion of the synovial membrane and the basic connective tissues and vessels (Haslett *et al*., 2002; Klippel *et al*., 2001), In addition to the perfusion of fibrin in the joint space, which leads to the formation of small fibrous nodes in the joint space. Both types of synovial cells A and B participate in synovial membrane hyperplasia, as early cell infiltration occurs in the synovial fluid represented by infiltration of lymphocytes and macrophage cells in the acute case of rheumatoid arthritis regardless of the duration of the disease and in this tissue B and T cells aggregate to form a focal group and form T cells are mostly CD4 + cells and CD8 + cells. CD4 + cells are present in a structure similar to the lymph follicle, and in some cases these cells have characteristics similar to their counterparts in the germinal centers, whereas the CD8 + cells spread in the connective tissue filtrate, and the expansion of the vessels in the synovial membrane is a distinct feature of the progression of inflammation. Synovial (Burmester and Pezzutto., 2003). One of the characteristics of secondary lymphoid follicles is the interaction of the germinal

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 4, 2021, Pages. 6756 – 6767
Received 05 March 2021; Accepted 01 April 2021.

center with B cells grouped around a network of branched follicular cells that occurs when the disease progresses. In the more advanced stages of inflammation, the presence of plasma cells, multinucleated giant cells, and a few mast cells are rare. The secondary endothelial layer in the synovial membrane of rheumatoid arthritis is distinguished by its ability to proliferate neoangiogenesis and tissue fibrosis, as hyperplasia of the synovial endothelium and lymphocyte filtration is observed and continuous proliferation in biopsy of persons with the disease in its early and late stages, which is more related to disease effectiveness (Klippel *et al*., 2001)

Table (1) shows the most important proteolytic enzymes in rheumatoid arthritis

Representative	Enzyme family
MMPS 1,2,3, and 9	**Matrix metalloproteinases (MMP)**
Cathepsins B, H and L	**Cysteine proteinases**
Elastase, plasminoger Activator, cathepsin G	**Serine proteinase**
Cathepsin D	**Aspartic proteinases**

The diagnosis

Rheumatoid arthritis is a chronic disease that is difficult to diagnose and separate from other diseases. Diagnosis depends on clinical signs, radiological results and a number of laboratory tests. The disease begins with the emergence of joint symptoms that usually include symmetrical joints and the progression of the disease is slow. Symptoms appear in a period of one week to a month and are severe in the beginning, injuries of the metacarpophalangeal joints and the joints between the proximal interphalangeal joints of the proximal interphalangeal joints are among the early clinical signs of the disease. The diagnosis can be based not only on clinical signs, radiological results, and a number of laboratory tests, but on an analysis of the fluid in the joint, which can be examined and other diseases ruled out. (Shaun., 2003 and Westwood *et al*., 2006).

Most patients suffer from non-specialized systemic symptoms that include fatigue, anxiety, mild fever and severe depression, and these symptoms exceed the typical clinical signs of the disease during a period of a week or a month, and these symptoms are in an increase or decrease, especially at the onset of the disease, and this leads to a delay in the diagnosis and from the basic characteristics that Diagnosis supports the occurrence of morning stiffness and the emergence of rheumatoid nodes as well as the rheumatoid factor and synovial fluid inflammation characterized by an increase in the number of polymorphic white cells, the results of radiographs of the joint, and erosion of affected joints (Shaun, 2003 and Nishimure *et al*., 2007), The diagnosis is based on an increase in the distinctive clinical signs and that the results based on a positive rheumatic factor test and a high erythrocyte sedimentation rate are not considered a diagnostic for rheumatoid arthritis, especially in elderly people (Kasper et al, 2005). In 1987, the American Rheumatology College developed a number of Criteria for classification of rheumatoid arthritis and the sensitivity of these

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 4, 2021, Pages. 6756 – 6767
Received 05 March 2021; Accepted 01 April 2021.

criteria in diagnosis is between 91-94% and their specificity is 89%. These criteria include:

1. Morring ctiffnes, in which there is stiffness in the joints and their surroundings, which lasts for an hour or more.
2. Inflammation of three or more joints.
3. Arthritis of the hands, as it includes at least one joint.
4. Symmetrical arthritis.
5. Rheumatoid nodules, which are nodes under the skin and surfaces of the extensor muscle or areas adjacent to the joint.
6. The appearance of rheumatoid factor (RF +) in the serum.
7. Radiographic changes, which include erosion of the bones adjacent to the covered rheumatoid joints.

The longer there are four or more of these criteria. Diagnosed for rheumatoid arthritis, and that the first four symptoms may appear during the first six weeks of infection, but the presence of two symptoms does not exclude the possibility of infection.

In the initial diagnosis of autoimmune diseases, a number of tests are conducted, some of which are not specialized, such as the total number of white blood cells, the erythrocyte sedimentation rate, the presence of the rheumatoid factor, as well as protein and electrolyte tests. Complementary tests may be performed in which the level of hormones is measured.

Figure (1) shows the effect of rheumatoid rheumatoiditis on the wrist joint (Shaun., 2003)

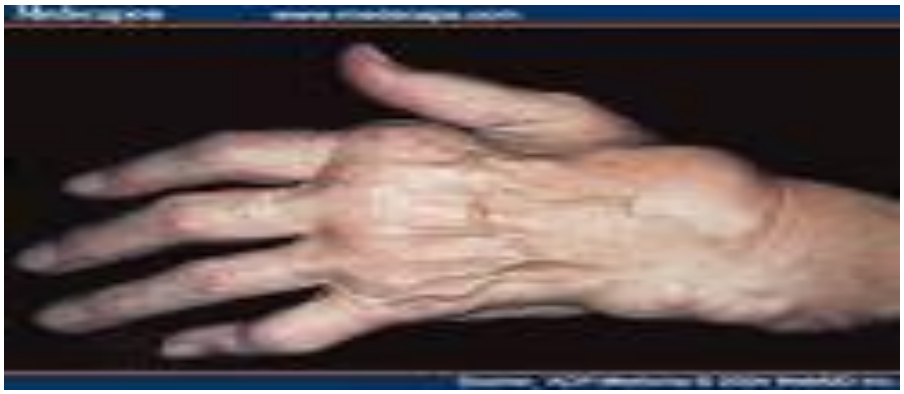


Figure (2) shows the effect of rheumatoid rheumatism on the hands (Shaun, 2003)

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 4, 2021, Pages. 6756 – 6767
Received 05 March 2021; Accepted 01 April 2021.

Materials and methods

Sample study

The study included (55) blood samples distributed to (40) blood samples belonging to patients attending Tikrit Teaching Hospital and private outpatient clinics. Their disease was diagnosed by doctors and specialists through a set of criteria described by the American Society of Joint Diseases (Arnett, 1988).

After the interview and a number of laboratory tests, these samples were counted for a group of patients aged between (20-60) years, and it consisted of (33) female samples and (7) male samples. As for the remaining samples of (15) samples, they are a group of healthy subjects that were considered a control group for the study and who did not show them the criteria described above. It consisted of (5) male samples and (10) female samples whose ages ranged between (20-60) years, and the studied samples (patients and healthy subjects) were confirmed that they did not have any chronic diseases such as cardiovascular disease and diabetes. A form that included age, gender, and required tests.

blood

The volume of blood drawn was about 10 cm3 from healthy and sick people, and it was divided according to the type of examination, as 5 cm3 was placed in plastic tubes with caps containing an anti-coagulant (EDTA), This is for the purpose of conducting tests for assigning blood groups, (blood group), hemoclobin concentration (Hb), and percentage of compacted red blood cells (PCV).

serum preparation

The serum was obtained by placing the remaining volume of the blood drawn, as in the previous paragraph, in a plastic tube with a closed cap, free of anti-coagulation, and leaving the blood at a temperature of 25 degrees until it coagulated, and then placed in a centrifuge for 10 minutes and the serum was withdrawn (the filter) By a Micro Pipette and placed in clean and sterile tubes and kept in a state of freezing at a temperature of 20 ° - and the serum was used for hormonal and biochemical tests.

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 4, 2021, Pages. 6756 – 6767
Received 05 March 2021; Accepted 01 April 2021.

Blood tests:

The first section of blood drawn and placed in tubes containing EDTA was taken for the purpose of the following checks.

Rheumatoid factor (RF) identification:

The method used in the test (Singer and Reinthaler, 1956) which relies on the occurrence of rapid clumping and left the serum samples and the test kit to reach room temperature before performing the test and transferred (50) microliters of the serum sample to the slide for the examination containing 6 sections, then we added (50) Microliter of test-specific antigen and mixed with serum with rapid spin for two minutes, after which the result was recorded by observing the clumping in front of a light source.

Blood group identification

The method of work

The method of work was carried out by means of the slide method, which is a quick method. I took a clean glass slide and put three separate drops of the patient's blood in it, then put a drop of antibody (A) with one of the drops and a drop of antibody (B) with the second drop and a drop of antibody (D) with The last drop (mixing with a wooden stick for each drop separately) and after a minute of time with simple shaking, a note is made whether there is a compaction or not, then the results are recorded (Power, 1989).

Packed cell volume (PCV)

Small capillary tubes, 75 mm in length and an internal diameter of about 1 mm, were used. Two-thirds of the capillary tube was filled with blood by means of the capillary property, then one end of it was closed by the manufactured clay. The tube was placed in a Micro centerfuge at 5,000 rpm for 5 minutes. The interval between the plasma column and the blood cells is then measured by a special measuring ruler or by the Heamato Crite Reader and the percentage of the volume of the compacted blood cells is read (Powers , 1989).

Hemoglobine determination (Hb):

The value of (Hb) was calculated by dividing the result (the value of P C V) by 3.3, because hemoglobin represents 1/3 the volume of the red blood cell (Powers, 1989).

statistical analysis:

The following statistical methods were adopted in analyzing the results of the current study:

1- Standard statistical method for finding the arithmetic mean, standard deviation, standard line, number and percentage.
2- T-test for comparison between two groups.

The results of the tests were considered significant at a probability level ($p < 0.05$).

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 4, 2021, Pages. 6756 – 6767
Received 05 March 2021; Accepted 01 April 2021.

Patient group

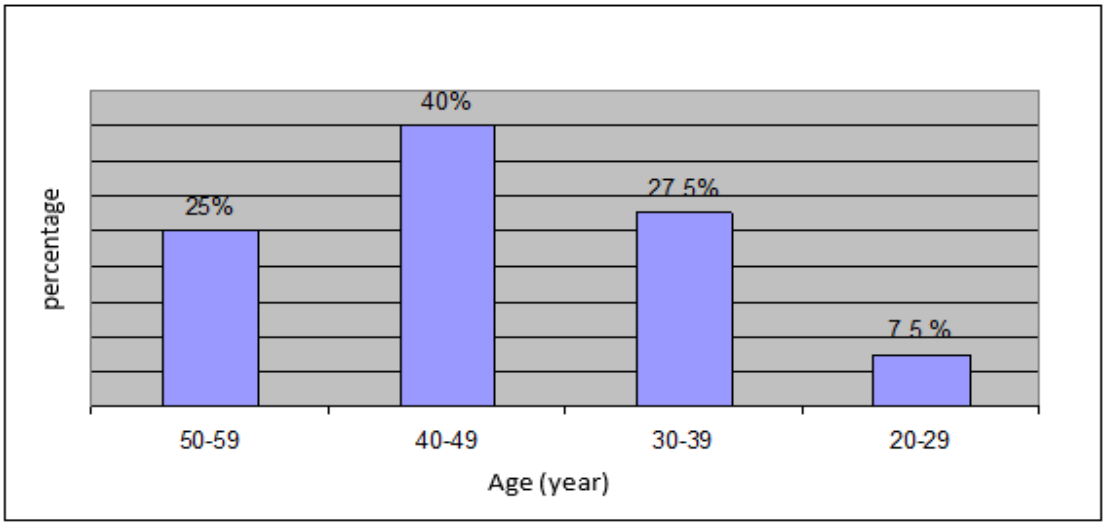
The number of patients with rheumatoid arthritis after conducting clinical and biochemical examinations and from both sexes (40) patients.

Discussion and Results

The relationship of injury with age

Figure (3) shows the percentages of patients with rheumatoid arthritis and according to different age groups. It is noticed that the highest incidence rate was in the age groups (30-39) years, followed by the group (50-59) years, and that the lowest incidence was in the group (20-29). Many studies have shown that the peak incidence is at ages ranging between (40-49) years, as the study (Kasper *et al*, 2005) indicated that the highest rate of infection is at ages ranging between (40-50) years, and the study indicated that The peak of infection is at ages ranging between (30-50) years, and this has been confirmed by other studies. And the prevalence of the disease increases with age until it reaches (70-85) years (Rubins *et al*., 2005).

Figure (3) Percentages of pneumonia cases in different age groups

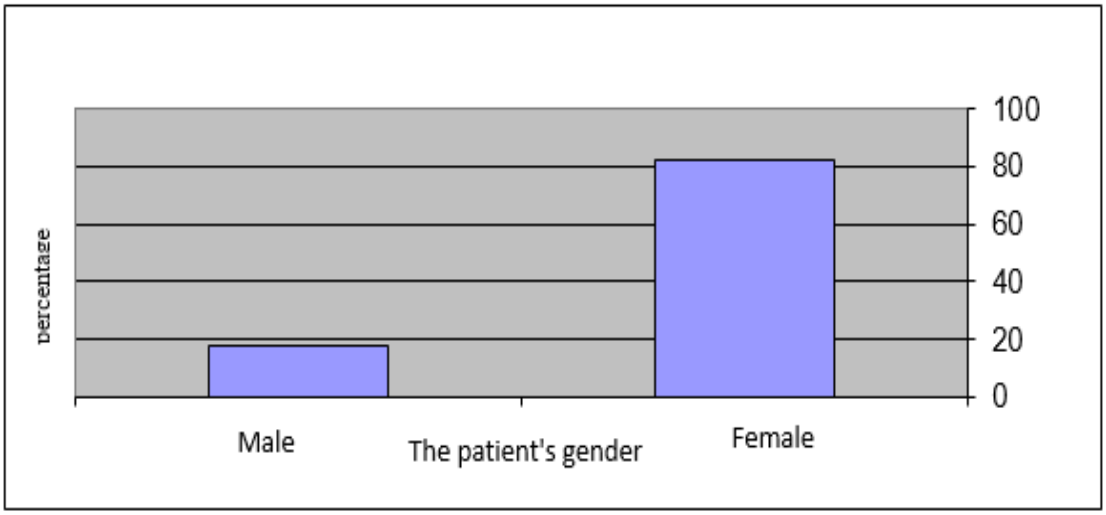


Impact of sex in rheumatoid arthritis

Figure (4) The percentage of the prevalence of rheumatoid arthritis according to the gender. A study showed that the rate of infection in females to males was (4: 1), while other studies showed (Mahon and Tice., 2006). The rate of infection between females to males is (1: 3) and that the difference in the rate of infection between the two sexes is close or equal at advanced ages. It was conducted in India and explained that this ratio was (1: 9) and that the difference in infection according to gender may be related to the effects. Hormonal, as it is believed that the estrogen hormone may be one of the factors encouraging the infection or it may be due to the genetic factors associated with the female chromosomes (X-chromosomes), which are x2 for females. They are approximately equal in both sexes.

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 4, 2021, Pages. 6756 – 6767
Received 05 March 2021; Accepted 01 April 2021.

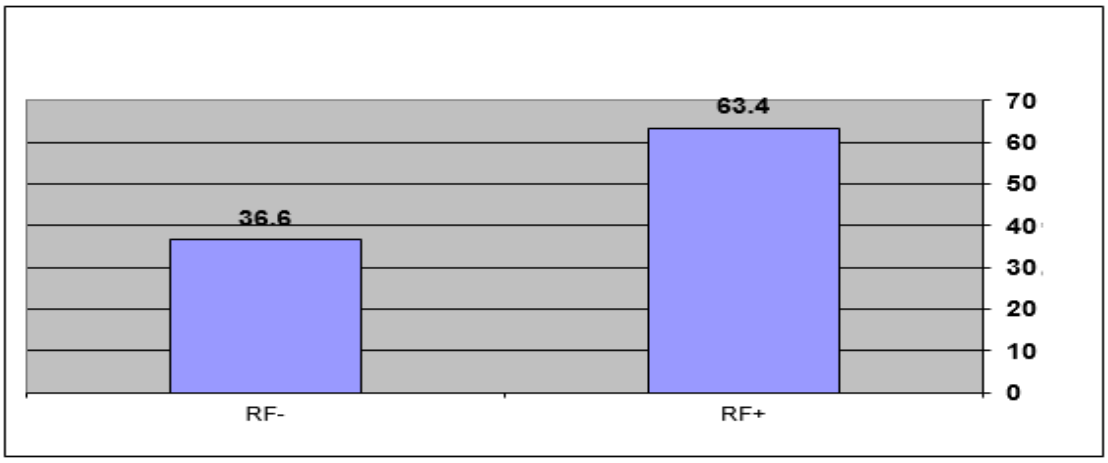
Figure (4) Percentage of rheumatoid arthritis by gender



Rheum factor examination:

The results shown in Figure (5) show the percentage of people with rheumatoid arthritis who showed a positive test for the presence of the rheumatoid factor in their sera, as the percentage reached (63.4%). The rate of positive infection ranged between 60-80% and it can be detected in the sera of relatives of people with rheumatoid arthritis, its presence is not considered a diagnostic of its progression, and the positive result of this examination is related to the appearance of the external symptoms and the formation of the rheumatic nodes (Rubins *et al*., 2005) and with progress Disease Patients may show a negative rheumatic factor test as a positive result of the test, and thus this examination is one of the important criteria in the diagnosis.

Figure (5) Percentage of rheumatic factor in patients with rheumatoid arthritis



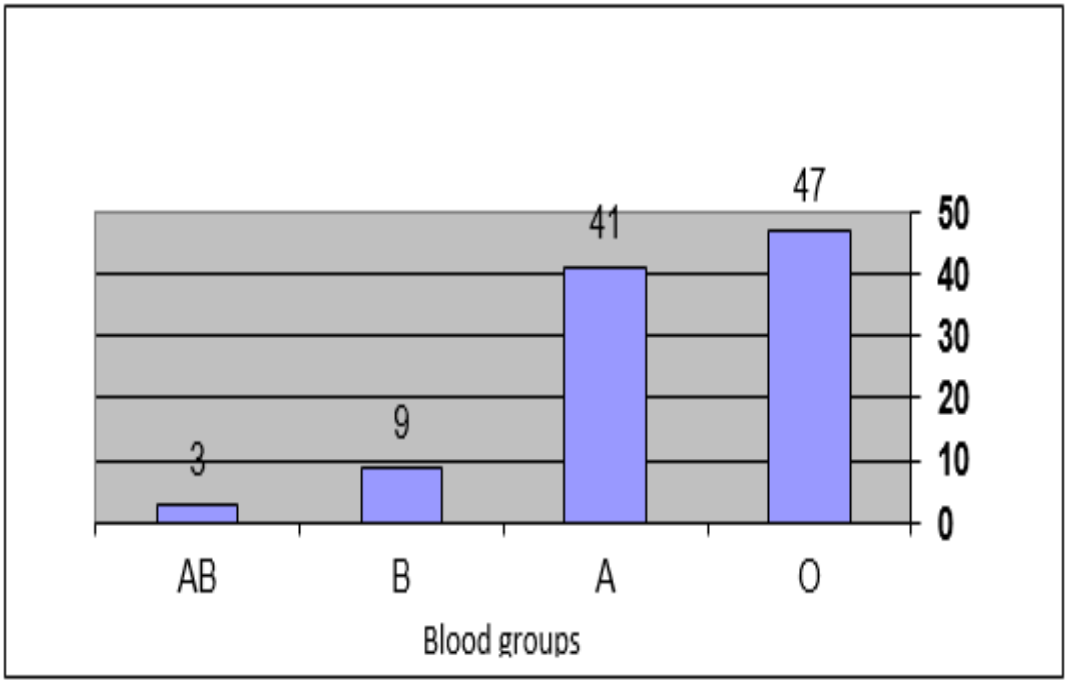
Relationship of injury with blood groups:

Figure (6) shows the percentage of rheumatoid arthritis in the different blood groups, as it is noticed that the highest rate of infection was in patients who had group O, at a rate (47%), followed by patients who had group A, at a rate (41%), and then

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 4, 2021, Pages. 6756 – 6767
Received 05 March 2021; Accepted 01 April 2021.

followed by patients who had group B. And by (9%), the lowest infection was among patients who had blood group AB, and by (3%). From these results, we find that the infection does not affect a specific group of patients, but rather according to the nature of the group's arrangement in healthy people. As for Rh-factor, it was within normal limits, and this indicates that the disease is not affected by it. Studies have confirmed (Singh and Hertello., 2005).

Figure (6) Percentage of rheumatoid arthritis according to blood group



The relationship of the disease to the hematological variants

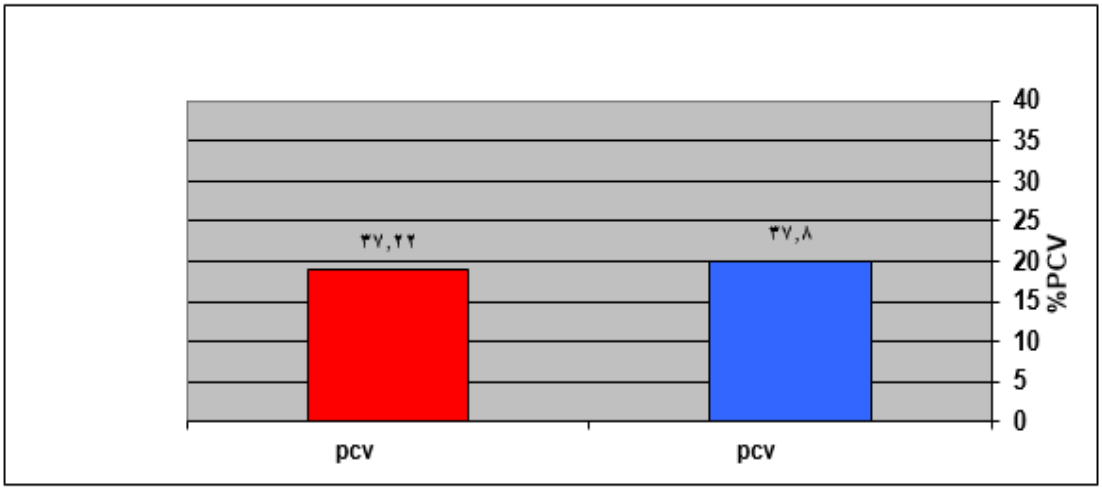
Compressed blood cell volume:

We notice through the stereoscopic (7) that there were no significant differences in the volume of compressed blood cells between the healthy group and the patient group at ($P < 0.05$) level. The percentage of compressed blood cells in the healthy group was 37.80 ± 2.0 compared to the patient group and it was 37.22 ± 0.83.

The results show an insignificant decrease in the volume of blood cells agglutinating in patients with rheumatoid arthritis than the healthy group, as it has a correlation with the hemoglobin concentration and the lower the volume of the compact cells, the hemoglobin concentration decreases with it. Reduced blood volume is an indication of anemia (Anemia) As a high value may be a sign of dehydration, a low value may be an indication of an increase in body water. The results show an insignificant decrease in the volume of blood cells compared with the control group. The reason for this is due to the increase in body water due to Uremia, which retains the largest amount of water, and the differences that exist between the values of the volume of blood cells compacting the disease.

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 4, 2021, Pages. 6756 – 6767
Received 05 March 2021; Accepted 01 April 2021.

Figure (7) shows the volume of compressed blood cells in the healthy and patient group

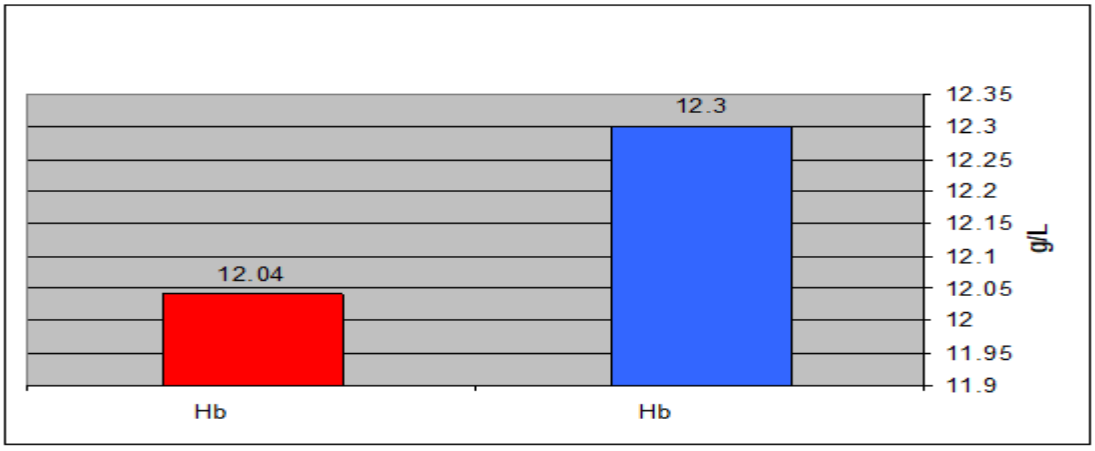


Hemoglobin concentration:

We find through Fig. (8) that there were no significant differences in hemoglobin concentration between the healthy group and the patient group at (P <0.05) level. We note in Table (5) that the average hemoglobin concentration in the healthy group was 12.30 ± 0.67 gm / 100ml, compared with the patient group, where it was 100 gm / 100ml12.04 ± 0.27.

This result shows an insignificant decrease in the level of hemoglobin in patients with rheumatoid arthritis than in the healthy group, and this is a decrease in the level of hemoglobin, which may be due to the breakdown of red blood cells or from the lack of red blood cells production as a result of a decrease in the level of the hormone Erthropoietin or a decrease in the bone marrow response Erthropoietin or as a result of the effect of anti-inflammatory drugs used in the treatment, or it may be due to a defect in the release of iron in the endothelial retinal tissues, and anemia is one of the common extroverted conditions associated with rheumatoid arthritis (Mahon and Tice *et al*., 2006).

Figure (8) shows the hemoglobin concentration in the healthy and sick group



Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 4, 2021, Pages. 6756 – 6767
Received 05 March 2021; Accepted 01 April 2021.

Reference

1. Aaran ,S.D.; Vandem Heen,K.L.; Naftel,S.A.;Lewis,M.J.and Rodger, M.A.(2003): Topical tetracaine prior to arterial puncture: randomized placebo-controlled clinical trial. Respire Med. 97(11):1195-1199.
2. Arnett ,F.C.;Edworthy,S.M. and Bloch,D,A.(1988). The american rheumatoid association 1987 revised criteria for the classification of rheumatoid arthritis. Arthritis Rheum,31:315-324.
3. Bessiere,C.M.;Sebbag,M.;Neuhauser,E.G.;Nogueira,L.;Vincent,C.Senshu,T.and Serre G.(2001).The major synovial targets of the rheumatoid arthritis-specific antifilaggrin autoantibodies are deiminated formas of the α and β-chains of fibrin.J.Immun.166:4177-4184.
4. Burtis ,C.A. and Ashwood E.R.(1994): Tietz Textbook of Clinical Chemistry 2n Ed, ISBN 0-72.
5. Noskov ,S.Y.; Berneshe,S.and Roux, B.(2004)Control of ion selectivity in potassium channels by electrostatic and dynamic properties of gabonyl ligands. Nature 431:830-834.
6. Majithia ,V.;and Geraci, S.A.(2007).Rheumatiod arthritis:diagnosis and management. Amirican. J. Med.120(11):936-9.
7. Klippel ,J.H.; Crofford,L.J.;Stone,J.H.and Weyand,C.M.(2001). Primer on the Rheumatic Disease. 12th ed., Arthritis Foundation , U.S.A.Atlanta.,pp.134-6.209-214.
8. Frank ,M.M.;Austen,K,F.;Claman,H.N.and Unanue,E.R.(1995).Samter's Immunologic Diseases:Vol. 11,5th ed.,Little,Brown and Company,Boston,pp:694-701.
9. Haslett ,C.; Chilvers, E.R.;Boon,N.A.;Colledge,N.R.and Hunter J.A.A.(2002). Davidsons Principles and Practice of Medicine 19th ed.Churchill Livingstone Edinburgh,963.1002-7.
10. Burmester ,G.R.and pezzutto,A.(2003).Color Atlas of Immunology Geore Thieme verlag stuttgrt medical Press.Germany .166-172.
11. Westwood ,OM.;Nelson, PN.and Hay, FC.(2006). Rheumatiod factors.Rheumatology 45(4):85-379.
12. Shaun R.(2003).Introduction to the rhaumated disease in:David.Dah Daniel .Federman Scientific American Medicine.WebMD.Ine new york.111:pps.1282-1287.
13. Nishimura, K.;Sugiyama,D.;Kogata,Y.(2007) Meta-analysis diagnosic accuracy of anti-cyclic citrullinated peptide antibody and rheumatoid factor for rheumatoid arthritis(PFD).Ann.Inter.Med.146(11):797-808.
14. Kasper ,D.L.;Fauci,A.S.;Longo,D.L.;Braunwad,E.;Hauser ,S.and Jameson, J.L.(2005). Harrisons Principles of Internal Medicinal16th ed.Mc Graw-Hill Companies,Inc. USA. V.2,1962-1975.
15. Singer ,F.and Reinthaler,M.(1956) Serum ferritin and iron uptake by peripheral blood leukocytes in patients with active rheumatoid arthritis.Clin Rheumatol.4:267-73.
16. Powers .L.W.(1989).Diagnostic Hematology: Clinical and technical principles. C.V. Mosby..U.S.A.60-80.

Annals of R.S.C.B., ISSN:1583-6258, Vol. 25, Issue 4, 2021, Pages. 6756 – 6767
Received 05 March 2021; Accepted 01 April 2021.

17. Robbins ,L.,Burckhardt,C.S.;Hannan,M.T.and Dehoratius,P.J.(2001).Clinical Care in the Rheumatic Disease.2nd ed.,American college of Rheumatology.Georgia .U.S.A.38-41,89-95.
18. Mahon ,C.R. and Tice,D.(2006).Clinical laboratory lmmunology pearson Education,Inc., New Jersey,147-151.
19. Singh ,A.K. and Hertello,P.(2005).The benefit of IV iron therapy in treating and anemia in patients with renal disease and comorbid cardiovascular disease. Nephrol. Nurs.J.32(2):199-208.