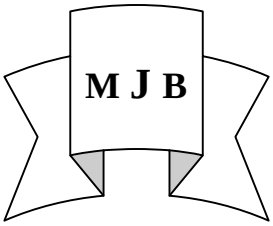
Immunology of Staphylococcal Human Persistent Pyuria

Ibrahim M.S.Shnawa and Baha,H .Al- amedi*
Dept. of Biology College of Science Babylon University. Hilla p.o box 4 IRAQ
*Public Health Laboratory Hilla .Babylon, IRAQ



Abstract

Fifteen *Staphylococcus aureus* isolates from human with persistent pyuria were diagnosed .Patient had high total serum protein (7.178 g/dl) , high serum (4.081 g/dl) and mucosal globulins concentration (0.072 g/dl] and lowered serum albumin globulin ratio (56.68%) in comparison to normal subjects.
These patients were with significant anti *S . aureus* specific agglutinins both at serum and mucosal secretions . Their peripheral and mucosal leucocyte migration inhibition (LIF) were significant (peripheral 0.5, mucosal 0.58) .Non significant anti *S. aureus* agglutinins and LIF were noted in blood and secretion of five immunecompromized, these five cases were showing low grade (specific antibody titre 40, LIF, peripheral 0.8, mucosal 0.75) immune responses due an immunocompromy. The involved immunodominant epitopes in these immune responses may be T dependent types that can activate either or both Th1,Th2 . For humoral arm direct B cell epitope maybe expected also .

الخلاصة

تم تشخيص خمس عشر مريض بالبيلة القيحية المستديمة المشتركة مع المكورات العنقودية الذهبية .تبين هؤلاء المرضى ارتفاع واضح في كل من البروتين المصلى الكلى والكلوبيولين المصلى والمخاطي وانخفاض في نسبة الالبيومين إلى الكلوبيولين المصلى .

واظهر هؤلاء المرضى ارتفاع نسبى في اللزائن المتخصصة بالمكورات العنقودية الذهبية في الأمصال وفى الكلوبيولين المناعي المخاطي .وكانت معاملات تثبيط هجرة الخلايا البيض من الدم المحيطي و الخلايا البيض المخاطية معنوية . وفى خمس مرضى آخرين لوحظ اختزال في مستويات اللزائن المتخصصة في المصل المخاط وكدا الأمر حصول معاملات تثبيط هجرة الخلايا البيض غير معنوية في الدم المحيطي وفى المخاط .
ويعزى دلك لانخفاض مناعي عند هؤلاء المرضى . ويمكن تفسير نوع الذرى المستضدية ذات العلاقة بهذه الاستجابات بأنها ذات صفة معتمدة على الخلايا التائية .

Introduction

Shnawa [1] and Al – Nasiri [2] have been reported *S . aureus* persistent pyuria (chronic and recurrent] in man . Mehdi [3] , studied the mucosal humoral immune responses in a group of uropathogens including *S . aureus* persistent pyuria in this area, and else where in the world [4] .

The objective of the present work was at investigation of humoral and cellular immune responses against *S . aureus* persistent pyuria in man .

Materials and methods

Fifteen immunoreactive and five immunocompromized persistent pyuria patient were investigated . From each of

which (15-20 ml) clean catch midstream urine & 5 ml blood with and without anticoagulant were collected [5, 6,7] . Ten normal human (control) subjects were sampled as for test group. The bacteriology and immunology of these samples were performed by standardized methodology [6 – 17] and were briefed in the followings :

The urine samples were processed for culture by direct quadrate streaking technique [6] as well as by indirect broth enrichment and quadrate streaking technique [7] . Growth on blood and MacConkey agar plates were checked. Pure isolates were identified by classical and miniaturized biochemical tests [6 – 7] . Whole cell somatic *S.aureus* antigens were prepared by benzelchonium chloride [8] . Cell free culture filtrate was made from 24 broth culture after centerfugation for 10 min. at 5000 RPM . Supernates were filtered through, 0.22 Millipore microfiltration unite of injection type . This preparation was used as sensitizer in leukocyte inhibition test . The leukocyte inhibition test was done by capillary method using peripheral blood leukocyte and mucosal leukocyte [9 , 12] . The immunoglobulin classes were determined by specific anti immunoglobulin partigen plates according to Mancini and Laurel [13] . The separation of mucosal immunoglobulin (MIG) was done by 6% PEG 6000 [14] . MIG was partially characterized as ; dialysis for three days using saline for two shifts and distilled water for one shift [15] , protein determination by biurate method [13 – 14] , slide agglutination , standard tube agglutination with and without 2ME treatment [9] .

Results

Bacteriology : the isolates were identified as *S . aureus* (table – 1)

Immunology : the female / male ratio was 11 – 9 . the age range of the patients were 21 – 70 years .

The total serum protein median concentration was 7.128 g/dl .The serum albumine to globulin ratio was 0.75 these values were generally higher than those of normal values .

The medians of immunoglobulin class concentrations were 1.577, 0.208 and 0.212 g/dl in comparison to.1.197,0.297and 0.295 and 0.294 g/dL for IgG, IgM, IgA. Of control normal subjects .

The *S.aureus* specific agglutinins in sera and mucosal globulins were with titres of 360 and 40 for the immunoreactive as well as 240 and 40 for the immunoc-ompromy patients . The ratio of systemic to mucosal titres were nine to one and six to one respectivety (table – 2) .

The relationships between the median concentrations of globulin in serum and mucosal and the specific anti *S.aureus* agglutinin titres were found to be of simple linear type as they were indcated by regression analysis (fig–1) , for Serum the regression analysis equation was ;Y = - 1055.81 + 33.1904 x , R= 0.673Fc = 14.9107 at p=0.01. and Ft = 8.28 at p = 0.01 while for mucosa , the equation was ; Y = - 7.447 + 54.647 x, R = 0.630. ,Fc = 11.902 at p = 0.01 and Ft = 8.2 at p = 0.01 . The LIF in immunnoreactive patients were : 0.5 and 0.58 in peripheral blovd levcocytes and mucosal leucocytes , respectively . In immunocompromy patients , they were 0.8 and 0.75 for blood and mucosal accordingly .

Discussion

The associated uropathogen was gram positive, cocci in clusters, salt tolerant, mannitol and coagulase positive, such characters are consistent with *S .aureus* (table – 1) as reported in previous studies in this area [1 ,2] .

Persistent *S* . *aureus* pyuria can be attributed to cell wall defective from , antibody coated *S* . *aureus* and/ or small colony vanants [2 , 18] . *S.aureus* contained an arry of virulence associated antigens like , protein A , leucocydins , haemolysins and coagulases [19] .

Whether the infections were of ascending , dessending haematogenens or lymphogenens origins . They will reachs cell population to produce communication signals , the quorum sensing signals and startrs their pathogenic effects [20] . Usually , uropathogens of their products generate costimulatory , signals for T cells at the level of antigen presentation . In case B cells responding to T dependent antigen, the critical costimulatory signals are then provided by T helpers cells . A situation in which B cells could functions as :

1. generate high affinity antibody .
2. to switch from IgM to other Igs.
3. Proliferate to preduce APC and memory cells .

The abovementioned response many be right for protein A of *S.aureus* . for polysaccharid or tiechoic antigenic epitopes T indepedent type one is dircetly mitogenic to B cells . CD14 however may functions as (pattern recogition receptors which activate macrophage . the B cell mitogenic properties of these *S.aurues* epitopes are not yet well characterized . On the other hand , epitope reacts with a variety of receptors integrins , lectins and others. Invivo , cytokines , inflammatory mediators cell a dhesion , cell interaction molecules from various accessory cells can participate in Type1 indeperdent B cell responses [21] .

Significant LIF results indicate that the LIF cytokines are produced by the activated lymphocytes , hence cell mediated immune responses are involved and

are derived from *S.aureus* through these states of infection uropathies [22 , 19] .

Superantigen of T dependent and T independent types are expected to play a role in the cellular immune responses to *S.aureus* persistent pyuria patients [21 , 23] .

The noted immunocompromized state may be due to Diabetes [24] or Aging [25 , 26] and chronic infections [26] . Significant F values were noted on regression analysis for concentrations and titers [27] .Thus , *S.aureus* induce mucosal and systemic as well as cell and humoral immune responses . Immunocompromy , however suppresses both humoral and cellular arms of the immune responses [25,26] .

Table 1 Characterization of staphylococcus

Characters	Conventional *S.aureus*	Epi 20
Gram reaction	+	+
Shape	Cocci	+
Arrangement	Culsture	+
Motility	-	-
Catalase	+	+
Oxidase	-	-
Coagulase	+	+
B haemolysis	+	+
Salt tolerance	7-100	7-10
Mannitol	+	+
Maltose	+	+

Table 2 The immunology of *S. aureus* persistent pyuria

Character	*S.aureus*	Control
Age range		
11-20	1	-
21-29	2	2
30-39	10	3
-40-49	3	3
50- 59	2	2
-60- 70	2	-
Serum total protein median g/dl	7.178	7.027
Scrum globulin median g/dl	4.081	3.492
Mucosal globulin median g/dl	0.072	0.02
Serum globulin / mucosal glob ratio	56.68	174.6
Albumin / globulin ratio	0.75	1.01
Median of Sp. Aggl titre	360	-
Median of sp. Aggl titre mucus	40	-
Median of sp. Aggl titre comp	240	-
Median of sp. Aggl titre mucn / comp	40	-
Median systmic LIF	0.5	-
Median systmic LIF / comp	0.8	-
Median mucosal LIF	0.58	-
Median mucosal LIF/ comp.	0.75	

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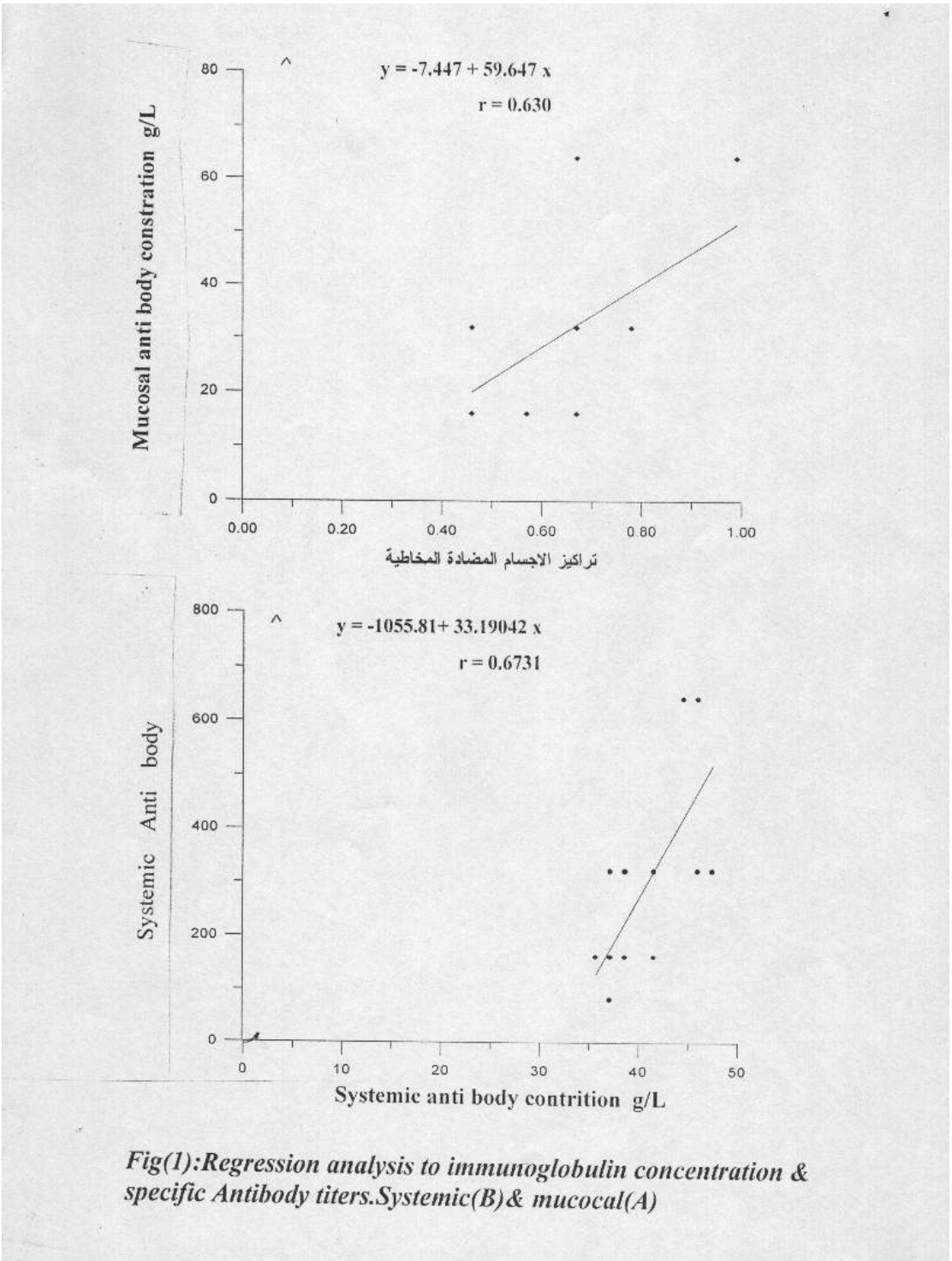

Fig(1):Regression analysis to immunoglobulin concentration & specific Antibody titers.Systemic(B)& mucocal(A)