

ORIGINAL ARTICLE

Bacterial Skin Colonization in Patients with Atopic Dermatitis/Eczema Syndrome

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ABSTRACT

From a total of 286 cases of AD patients, 94.4% and 86.36% yielded a positive cultures of eczematous lesions and healthy area of AD skin. Twenty various bacterial types and a total of 959 and 744 isolates were identified from each of above areas respectively. *Staph. aureus* was the predominant bacterial agent isolated from 60.48% of eczematous lesions, while *Staph. epidermidis* was predominantly from 57.34% of healthy areas of AD patients ($P < 0.001$). The bacterial numbers ranged from $(0.02-92.0) \times 10^5 \text{ cell/cm}^2$ in eczematous lesions and $(0.11-23.0) \times 10^3 \text{ cell/cm}^2$ in healthy areas of atopic skin ($P < 0.001$).

Key words: Atopic dermatitis (AD), bacteria, patient skin.

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INTRODUCTION

Atopic dermatitis (AD) is a chronic, pruritic eczematous disease that early always begins in infancy and childhood and follows a remitting / flaring course that may continue throughout life [1]. It develops as a result of a complex interrelationship of environmental, immunologic, genetic and pharmacologic factors [2]. It may be exacerbated by infection, psychological stress, seasonal of climate changes, irritants, and allergens [3]. The rapid rise in prevalence of AD is thought to be primarily related to changes in our environment. A number of factors can trigger AD, including irritants foods, aerollergens, and infection [4].

The skin of patients with AD is characterized by higher levels of colonization by aerobes [5]. Significantly higher colony forming units per surface unit could be isolated from clinically normal skin of patients with AD than from corresponding skin sites in normal controls [6]. Acute exudative lesion yielded a mean of 107 colony forming units (CFU)/cm². Thus, the atopic skin must provide a favorable environment for the colonization and multiplication of aerobic bacteria [5]. Coagulase-positive staphylococci (*Staph. aureus*), which are usually not found on normal skin, accounted for the majority of CFU isolated from AD skin [7]. *Staph. epidermidis* was the predominant organism isolated from the clinically uninvolved skin of AD patients, whereas *Staph. epidermidis* was second to *Staph. aureus* found in lesional skin [5]. It is generally accepted that the normal resident skin flora includes micrococci, staphylococci (e.g. *Staph. epidermidis* and *Staph. capitis*), corynebacteria, propionibacteria (e.g. *pr.acnes* and *pr.granulosum*) and *Acetino bacter*. An aerobes out number aerobes by a factor of more than 10, the latter containing most of the pathogenic organisms [4].

There is no previous study in Iraq - in especially - detailed bacterial colonization of atopic skin, so this study was found necessary to determine the bacterial types and numbers of each eczematous lesions and healthy areas from patient's skin with atopic dermatitis.

MATERIALS AND METHODS

Patients: A total of (286) patients suffering from atopic dermatitis syndrome in various age groups and both sexes were included in this study. The patients attending the out patients of dermatology department of fourth main hospitals in Basrah providence (out patients based study), and AD diagnosed under supervision of dermatologists based on a criteria of [8,9,10]. The study was carried out during period from November 2003 to July 2005.

Primary isolation: Brain Heart Infusion Broth (BHIB) saturated skin swabs from both eczematous lesions and healthy area were cultured on primary isolation media: Blood Agar Base BAB (Oxoid), MacConkey Agar MA and Nutrient Agar NA (Himedia) and then incubated at 37°C for 24-48 hrs

aerobically [11]. Samples that cultured on Chocolate Agar (CA) in addition to NA were incubated under CO₂ in candle jar at the same temperature and period time mentioned above [11].

Bacterial count : Enumeration of a total bacterial counts per cm² of the same above sites were carried by using a serial dilution technique of Nutrient Broth (HiMedia) and then culturing on NA[11].

Identification techniques: Four types of Api-technique (bioMerieux, France) as a rapid identification system were used for identification the various bacterial isolates based on enclosed instructions of supplied company:

Api Staph: identification system for staphylococci.

Api 20 Strept: identification system for streptococci.

Api 20 E: identification system for Enterobacteriaceae and other Gram-negative rods.

And Api 20 A: identification system for anaerobes.

Statistical analysis : Chi-Square test and ANOVA test were carried by using computer program , SPSS . ver.11.

RESULTS

Table (1) illustrate all bacterial types isolated from eczematous lesions and nearly healthy areas. The total numbers of positive cultures (270,247) cases from 286 studied AD cases in percentages (94.4, 86.36) from eczematous and healthy area respectively. In general, twenty bacterial types were isolated from both area separately and (959,744) isolates with isolation ratio (3.35:1, 2.6:1) isolates: case were identified in each above area respectively. The percentages of bacterial occurrence in eczematous lesions and healthy areas respectively were as follow: (P< 0.05) : *Staph. aureus* (60.48, 17.48), *Staph. epidermidis* (17.13, 57.34), *Staph. xylosus* (2.79) in each, *Staph. saprophyticus* (5.24, 10.48), *Staph. capitis* (2.79) in each, *Staph. hominis* (22.37, 9.44), *Strept.pyogenes* (17.13, 9.79), *Strept. Faecalis* (23.07, 17.83), *Strept. Mutans* (14.68, 9.44), *E.coli* (25.52, 33.21) *Enterobacter sp.* (5.59, 17.83), *Klebsiella sp.* (3.14, 1.39), *Acinetobacter sp.* (5.59, 3.49), *Proteus sp.* (5.94) in each, *Pseudomonas aeruginosa* (17.48, 5.59), *Propionibacterium acnes* (19.58, 3.49%), *Pr.granulosum* (20.27, 18.53), *Haemophilus influenzae* (21.32, 11.53), *Bacteroid sp.* (18.18, 3.84), and *Corynebacterium sp.* (26.92, 17.83).

Table (1): Illustrate bacterial types isolated from eczematous lesions and healthy areas of AD patients (P< 0.05)

Bacterial types	No. of cases (%) from eczematous lesion	No. of cases (%) from healthy area
<i>Staph. aureus</i>	173(60.48)	50 (17.48)
<i>Staph. epidermidis</i>	49 (17.13)	164 (57.34)
<i>Staph. xylosus</i>	8 (2.79)	8 (2.79)
<i>Staph. saprophyticus</i>	15 (5.24)	30 (10.48)
<i>Staph. capitis</i>	8 (2.79)	8 (2.79)
<i>Staph. hominis</i>	64 (22.37)	27 (9.44)
<i>Strept.pyogenes</i>	49 (17.13)	28 (9.79)
<i>Strept. Faecalis</i>	66 (23.07)	51 (17.83)
<i>Strept. Mutans</i>	42 (14.68)	27 (9.44)
<i>E.coli</i>	73 (25.52)	95 (33.21)
<i>Enterobacter sp.</i>	16 (5.59)	51 (17.83)
<i>Klebsiella sp.</i>	9 (3.14)	4 (1.39)
<i>Acinetobacter sp.</i>	16 (5.59)	10 (3.49)
<i>Proteus sp.</i>	17 (5.94)	17 (5.94)
<i>Pseudomonas aeruginosa</i>	50 (17.48)	16 (5.59)
<i>Propionibacterium acnes</i>	56 (19.58)	10 (3.49)
<i>Pr.granulosum</i>	58(20.27)	53(18.53)
<i>Haemophilus influenzae</i>	61 (21.32)	33 (11.53)
<i>Bacteroid sp.</i>	52 (18.18)	11 (3.84)
<i>Corynebacterium sp.</i>	77 (26.92)	51 (17.83)
No. of isolates	959	744
Average (isolate: case)	3.35:1	2.6:1
No. of -ve growth cultures	16 (5.59)	39 (13.63)
No. of +ve growth cultures	270 (94.4)	247 (86.36)
Total no. of cultures	286	

Table (2) illustrated modes of isolation of bacterial types from both areas of AD patients. It has been found that the double bacterial agents was predominant in each eczematous lesions and healthy area in percentages (44.11, 41.29) respectively followed by other modes, with significant differences between modes of isolation (P< 0.05).

Figure (1): showed distribution of bacterial types in eczematous lesions and healthy areas of skin of AD patients as previous described.

The means of bacterial number measured for studied areas of AD patients showed in figure (2). The bacterial numbers ranged from (0.02-92.0) X 10⁵ cell (CFU)/cm² in eczematous lesions and (0.11-23.0) X 10³ cell (CFU)/cm² in healthy areas with very highly significant differences between means of bacterial numbers within the same areas and between each studied area (P< 0.001).

Table (2): Illustrate modes of isolation of bacterial types from eczematous lesions and healthy areas of AD patients (P< 0.05)

Mode of isolation	No. of cases (%) from eczematous lesions	No. of cases (%) from healthy areas
Single	38 (14.07)	31 (12.55)
Double	75 (44.11)	102(41.29)
Third	22 (12.94)	19 (7.69)
Fourth	19 (11.17)	61 (24.69)
Fifth	59 (34.7)	17 (6.88)
Sixth and over	57 (33.52)	17 (6.88)
No. of + ve growth culture (from total No. of modes)	270 (94.4)	247 (86.36)
No. of - ve growth culture	16 (5.59)	39 (13.63)
Total no. of cases	286	286

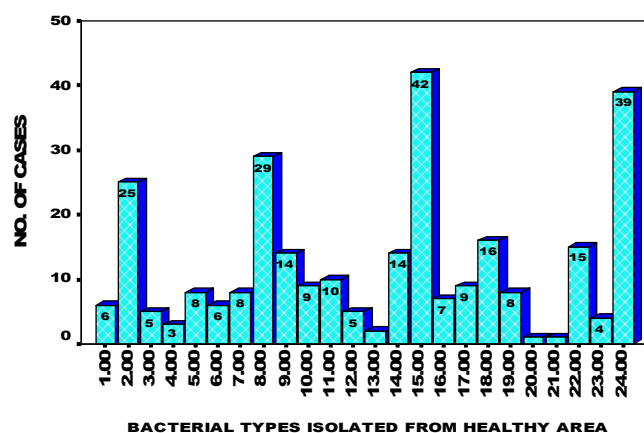
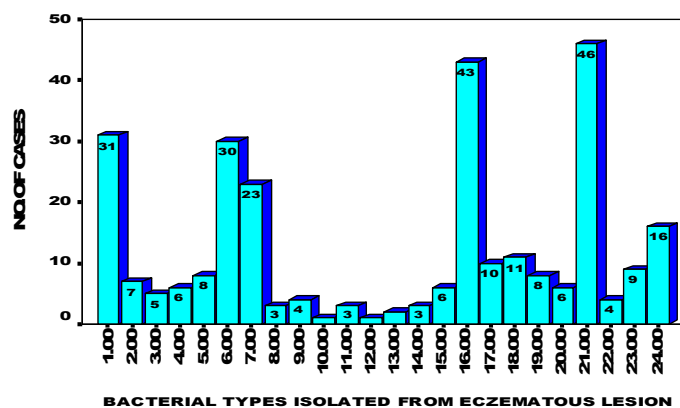


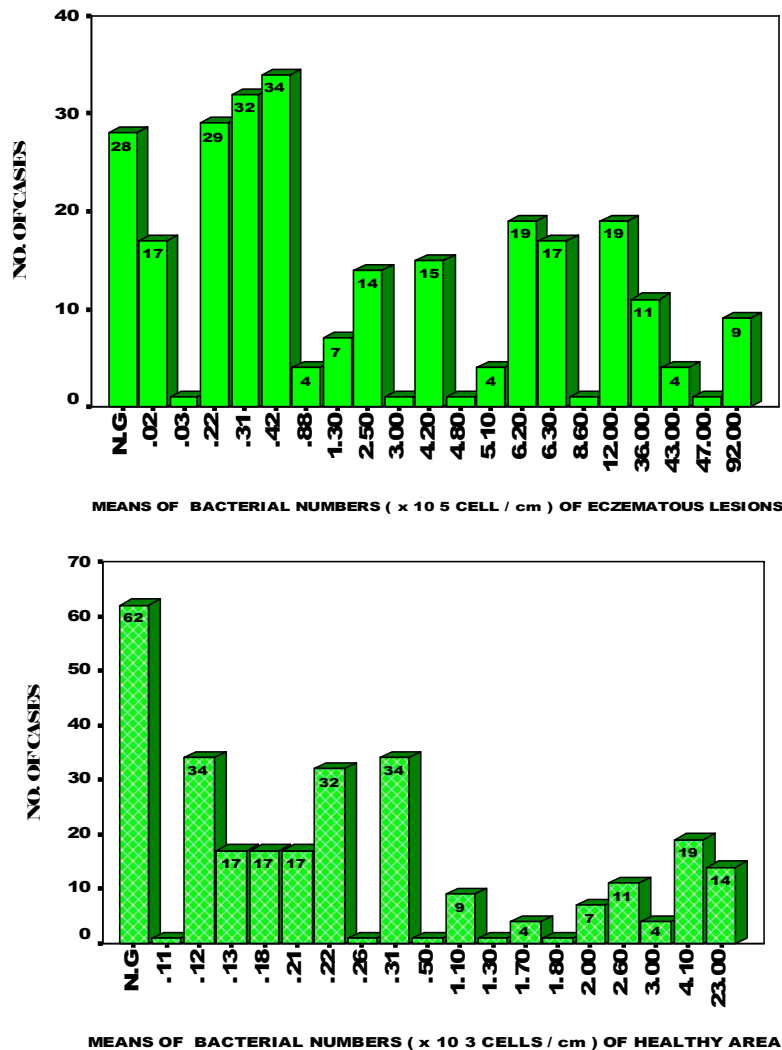
FIGURE 1: Illustrated Isolation Modes Of Bacterial Types From Eczematous Lesions (Above) & Healthy Area (Below) Of Ad Patients Skin . (P< 0.001)

Bacterial Types : 1 :*Staph aureus* , 2 : *StaphEpidermidis* , 3: *Staph Xylosum* , 4 : *Staph saprophyticus* , 5 : *Staph capitis* , 6 : *Staph hominis* , 7 : *Strept pyogenes* , 8 : *Strept faecalis* , 9 : *Strept mutans* , 10: *EcColi* , 11 : *Enterobacter Sp.* , 12 : *Klebsiella Sp.* , 13 : *Acinetobacter Sp.* , 14: *Proteus Sp.* , 15: *Ps Aeruginosa* , 16: *Pr acnes* , 17: *Pr Granulosum* , 18: *H Influenzae* , 19: *Bacteroid Sp.* , 20 : *Corynebacterium Sp.*

Isolation Modes : (As Show In Figures) Depend On Above Numbers

1 : 1 , 2: 2 , 3: 1&2 , 4 : 1&2&7 , 5 : 1&2&8 , 6: 1&7 , 7 : 1&9 , 8 : 2&8 , 9 : 2&4 , 10 : 2&6 , 11 : 2&7 , 12: 2&8 , 13 : 2&9 , 14: 2&10 , 15: 2&10&11&17 , 16: 1&10&17&19&20 , 17: 7&11&13&16&20 , 18 : 4&6&9&10&14&18&20 19 : 3&5&8 , 20: 6&9&10&13&14 , 21: 1&6&8&15&16&18 , 22: 10&15&18&20 , 23 : 12&17&19&20 , 24: NO GROWTH.

Figure 2 : Illustrated Means of Bacterial Numbers Measured For Eczematous Lesion And Healthy Area Of Ad Patients . (P < 0.001)



DISCUSSION

Our results revealed that *Staph. aureus* was the predominant bacterial agent isolated from (60.48%) of eczematous lesion, while *Staph. epidermidis* was predominantly from (57.34%) of healthy area of AD patients. And we evidenced highly prevalence /or occurrence of bacterial types in each studied areas.

Our knowledge concerning the complex interaction between microbes ad skin inflammation of atopic dermatitis has improved dramatically and today the Gram-positive bacterium, *Staph. aureus* is recognized as an important triggering factors for the maintenance of skin inflammation and acute exacerbations of the genetically determined skin disease atopic dermatitis [12,13].

Recent studies confirmed results of our study such as Brook, et al. (1996) that found 36%, 20%, and 44% of aerobic, an aerobic, and mixed respectively isolated from AD patients, these bacteria were *Staph*

aureus, group A-streptococci, *E.coli*, *Peptostreptococcus*, *Prevotella*, *Porphyromonas sp.* And *Fusobacteriu*[14].

Others demonstrated that the colonization density of eczematous lesions can reach 10^7 CFU/cm² [15] and the skin of 100% of AD patients may colonized with *Staph. aureus*, up of 65% of all *Staph. aureus* strains isolated from lesional skin have been sown to produce exotoxins with superantigenic properties [16]. Also Heaton, et al. (2003) illustrated that the incidence of *Staph. aureus* colonization on the skin of AD patient is approximately 90%[17].

Many modern studies illustrated the factors whose relevance to the increased colonization of atopic dermatitis skin with bacteria -in general- and with *Staph. aureus* -in especially-such as produce of exotoxins [4,18], ability of bacterial types to adherence with host cells of atopic skin [19]. pH values were shifting toward alkalinity with adherence of *Staph. aureus* to human keratinocytes being highest at pH= 7-8 [20]. And extracellular lipids of stratum corneum of epidermal layer of skin, the quantitative and qualitative changes in lipid composition could result in diminished antibacterial activity [21].

CONCLUSION

There are a highly occurrence and variation of bacterial types in eczematous lesions and healthy areas of skin of AD patients, with highly numbers of bacteria in the first above area in comparison with the second that indicate presence of highly infections in the lesional skin. Also *Staph. aureus* was the predominant bacterial types in eczematous lesions of AD skin.

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