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Evaluation Of Risk Factors For Dermal Infections with *Staphylococcus aureus* and Methicillin Resistant *Staphylococcus aureus* Among Sheep In Diyala Governorate, Iraq

Zainab Bressam Fajer[1], Ali Ibrahim Ali Al-Ezzy[2], Ahmed H. AL-Zuhairi[1]

University of Diyala, College of Veterinary medicine, Department of Medicine[1]
University of Diyala, College of Veterinary medicine, Department of pathology[2]

Abstract

Background: *Staphylococcus aureus* is one of the dominant pathogenic bacteria among skin infections in human and animals.

Aims: To evaluate the risk factors of dermal infections with *S.aureus* and methicillin resistant *S.aureus* **(**MRSA)

Methods :standard microbiological procedures were used for isolation of *S.aureus* and MRSA

Results : No correlation reported between breed and *S. aureus* infections among sheep. Significant correlation reported between breed and MRSA infections on sheep. No correlation was reported between flock size and *S. aureus* infections among sheep. No significant correlation reported in the middle of flock size and MRSA infections among sheep. No significant correlation reported amongst season ,*S. aureus and* MRSA infections in sheep. No significant correlation reported between introducing new sheep to the flock, *S. aureus* and MRSA infections among sheep. probability for infection of wound with *S. aureus* was 0.831time. the risk estimate to get infection with *S. aureus* due to wounds was (1.133) time compared with (0.942) intact skin .The risk estimate to get infection with *MRSA* for sheep suffered from wounds was (0.689) time compared with (1.234) for infection that reported among sheep with intact skin.

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The risk estimate to get infection with *S. aureus* due to abscess was (0.159) time compared with (1.647) for intact skin. The risk estimate to get infection with MRSA for sheep suffered from abscess was (0.780) time compared with (1.122) for intact skin. The risk estimate to get infection with *S. aureus* due to dermatitis was (0.865) time compared with (1.042) for healthy skin. risk estimate to get infection with MRSA for sheep suffered from dermatitis was (1.721) time compared with (0.880) for infection that for intact skin. The risk estimate to get infection with S. aureus due to abrasions was (17.448) time compared with (0.634) healthy skin. The risk estimate to get infection with MRSA for sheep suffered from abrasions was (2.525) time compared with (0.883) for intact skin.

Conclusions:

Abrasions represent major risk factor for getting *S.aureus* and MRSA infection in sheep. The breed, flock size, season, wounds, abscess, dermatitis represent minor risk factor for getting *S.aureus* and MRSA infection in sheep

Key wards : Staphylococcus aureus ,methicillin resistant staphylococcus aureus, skin ,sheep

Introduction;

Staphylococcus aureus is infectious pathogen that causes several diseases ranging from skin infections to necrotizing pneumonia, bacteremia, and life-threatening sepsis **[1]**

Gram-positive cluster-forming, spherical cells 0.5-1 micrometer in diameter, non-motile, non-spore forming, glucose and mannitol fermenter, catalase and coagulase positive, smooth golden yellow colonies; aerobically or anaerobically (facultative), able to raise at variety 15-45 C, even at 15% Sod. Chlorides and stay alive over dehydrated atmosphere from days to seasons **[2]** .While MRSA a group of *S. aureus*, natively different as former strain by methicillin compounds resistant, developed naturally or picked up horizontally through biomarker gene transfer, thought responsible for several difficulty in treating infections **[3]** .MRSA acquired in three types (HA-MRSA),

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(CA-MRSA) and (LA-MRSA) depending on its transmissions through human population and genetic fragments of SCCmec **[4]**. MRSA emergence in 1960s, leads to great illness, high mortality with augmented cure prices **[2, 5]**.

Sex, age, climate, family, economic, society, education, jobs, hospitalization, pets, environments, rural/urban demographic variations were Factors affect distribution, investigated around planet by biochemical, molecular, studies to evaluate risk estimations by many authors and researchers **[6]**

Innate sex bias toward female protection against *S. aureus* skin infection due estrogen effect was reported by **[7]** through Murine patron experiments. Epidemiological expanding related to Global warming and weather changes as an occurrence due to several matters, containing regularity of hotness surfs, hurricane streams cruelty, fall configuration variations, overflowing, and shore corrosion**[8]**. Several virulence factors allow adhering to surface, invading, and avoiding immune system, later producing toxic damaging the host tissues**[9]**.

Materials and Methods:

Study area and study population

This study was conducted on newborn to less than 1 year old Iraqi calves , living in the Baqubah city - Diyala province 33°45'34.71"N; 44°36'23.97"E ,Northeast[10-15].

Samples : A total of 75 skin swaps collected from south east distracts of Diyala governorate (Baladruze, Baqubah, Kanaan and Buhruz) in Iraq from 1st October 2021 to the end of February 2022, involving sheep suffered from variety of infected skin lesions (wounds, abscesses, dermatitis, abrasions) recording; sex, breeds, season, flock size, introducing of new

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sheep, to detect *S. aureus*, MRSA and estimate risk factors.

Employing traditional laboratory methods (Mannitol, Gram stain, Catalase, Coagulase, Nigrosin Capsule staining, and DNase) in addition to confirmatory techniques through fast rapid VETEK2 system, later well-known molecular genes assay (conventional PCR), Which applied for detection of *S. aureus* using the specific primer(Staur 4, 6)

Staur 4	**5'-ACGGAGTTACAAAGGACGAC-3'**
Staur 6	**5'-AGCTCAGCCTTAACGAGTAC-3'**

While MRSA verified innately via *mec*A gene primer;

Methicillin Resistant Gene	***mec*A**	***mec*A-F**	**162bp**	**5-TCCAGATTACAACTTCACCAGG-3**
		*mec*A-R		**3-CCACTTCATATCTTGTAACG-5**

Ethical consideration:

This study conducted according to the principles of Helsinki declaration. A full explanation of the purpose of this study to all owners before starting. Dully filled consent form obtained from all owners who agree to participate in the study. Approval of an ethical review committee of pathology department, college of veterinary medicine, Diyala University, Iraq, taken before initiation into the work[13, 16-26].

Statistical Analysis:

the Statistical Package for the Social Sciences windows version 17 (SPSS, Armonk, NY: IBM Corp)**[27, 28]** was used . Pearson's chi-square and Pearson's correlation coefficient

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was utilized for the correlation between the changeable of 2 test. P value of ≤ 0.05 and ≤ 0.01(2-tailed) were set to be statistically significant [29, 30].

Results:

Isolation rate :

As shown in table (1), *S. aureus* was isolated from 46 of 75 sheep skin lesions, rated (61.33%) while MRSA was 14/46, (30.43%) among positive isolates representing (18.67%) from total samples 14/75 according to methicillin resistance among Muller Hinton medium which confirmed by conventional PCR using *S. aureus* 23s RNA gene sequence specific primer (staur4 and staur6); (Figure 1) and S. aureus (*mec*A gene), (Figure **2**). agreed by Vitek 2 system.

Table (1): Isolation Rate of *S. aureus* and MRSA from Sheep

Typical Growth features of *S. aureus* on Mannitol salt agar	Methicillin *Resistant S. aureus*		Total
	Negative	Positive	
Negative	29(38.67%)	0(0%)	29(38.67%)
Positive	32(42.67%)	14(18.67%)	46(61.33%)
Total	61(81.33%)	14(18.67%)	75(100%)
χ^2	10.852		
P value	0.001		
R	0.380		
P value	0.001		
Likelihood Ratio	15.669		
P value	0.000		

Significant difference ($\chi 2=10.852$; p value =0.000) was reported between samples regarding the type of isolates *S. aureus* versus MRSA with significant likelihood ratio (p value =0.000)

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and correlation between the positive *S. aureus* positive samples to be MRSA (p value =0.000).

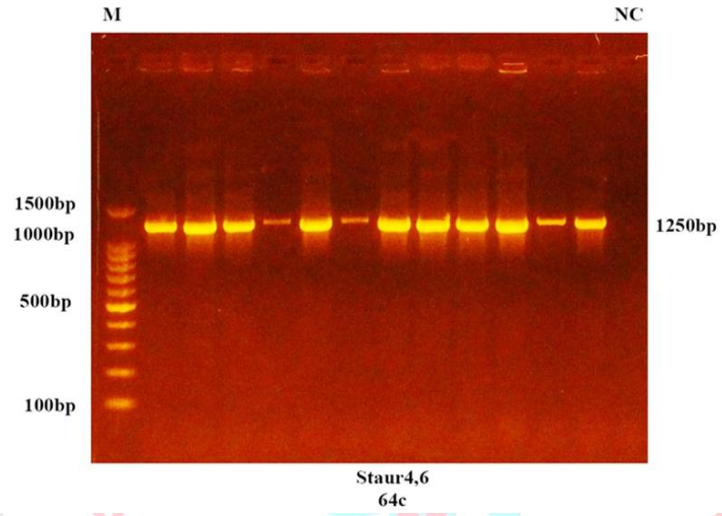

Figure (1): Amplification for staur primers 4&6 (1250bp) by conventional PCR for *S. aureus* recovered from skin lesions of sheep .The amplified DNA product was fractionated,
NC: Negative control.

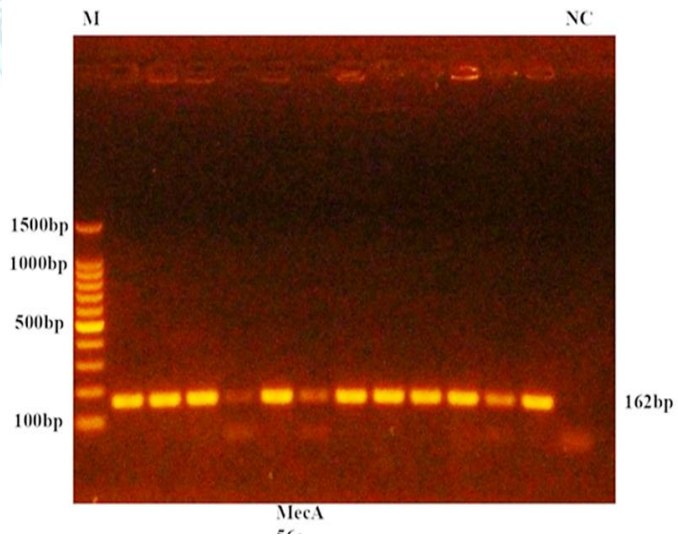

Figure (2): Amplification MecA (162bp) by conventional PCR for *S. aureus* recovered from skin lesions of sheep, the amplified DNA product was fractionated.

NC: Negative control.

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A-Sex factor: As shown in table (2), *S. aureus* and MRSA infections were concentrated among females 31/75, (41.33%) versus 9/75, (12%) for MRSA, while males infection with *S. aureus* represent only 15/75 (20%) versus 5/75, (6.66%) of MRSA.

Female probability to infect with *S. aureus* or (odds ratio) was 1.521 time greater than males, risk estimate for male to get infection with *S. aureus* was (0.740) time compared (1.126) for females, female probability to infects with MRSA or (odds ratio) was (1.438) time greater than males, risk estimate for male to get infection with MRSA was (0.780) time compared with (1.122) for females.

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Table (2): Sheep Sex As A Risk Factor For Infection With *S. aureus* and MRSA

Type Of Isolates From Skin Lesions Of Sheep					
Sex	*S. aureus*		MRSA		
	Positive	Negative	Positive	Negative	Total
Female	31(41.33%)	22(29.33%)	9(12%)	44(58.67 %)	53(70.67%)
Male	15(20%)	7(9.33%)	5(6.67%)	17(22.67 %)	22(29.33%)
Total	46(61.33%)	29(38.67)	14(18.67 %)	61(81.33 %)	75(100%)
Odds ratio for sex (Female/Male)	Value	95% CI	Value	95% CI	
	1.521	0.532- 4.348	1.438	0.421-4.910	
Risk estimate for male	0.740	0.344-1.595	0.780	0.347-1.755	
Risk estimate for female	1.126	0.845- 1.500	1.122	0.737-1.708	

B- Breeds:

There was two main breeds, As shown in table (3), *S. aureus* and MRSA infections were concentrated among mixed breed 37/75 rated (49.33%) versus 9/75,(12%) for Awassi, on the other hand, infection with MRSA was reported only among mixed breed, 14/75, (18.66%).

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Table (3): Sheep Breed As A Risk Factor For Infection With *S. aureus* and MRSA.

Breed	Type Of Isolates From Skin Lesions Of Sheep				
	S. aureus		MRSA		
	Positive	Negative	Positive	Nega-tive	Total
Awassi	9(12%)	6(8%)	0(0%)	15(20%)	15(20%)
Mixed	37(49.33 %)	23(30.67 %)	14(18.66 %)	46(61.33 %)	60(80%)
Total	46(61.33 %)	29(38.67)	14(18.67 %)	61(81.33 %)	75(100%)
χ^2	0.014		4.303		
P value	0.906		0.038		
R	0.014		0.240		
P value	0.907		0.038		
Odds Ratio for	Value	95% CI	Value	95% CI	
breed (Awassi / Mixed)	1.072	0.337-3.410		ND	
Risk estimate for Awassi	1.057	0.420-2.660		ND	
Risk estimate for Mixed	0.986	0.780-1.246	0.754	0.653- 0.870	

Neither Significant difference ($\chi 2$=0.014; p value =0.906), nor correlation (R= 0.014; p-value =0.907) reported between breed via *S. aureus* infections among sheep, Significant difference ($\chi 2$=4.303; P value = 0.038), Correlation (R = 0.240; p-value = 0.038) reported amid breed and MRSA infections on sheep, probability of breed (Awassi / Mixed), for infection with S. aureus or (odds ratio) was 1.072 time, Awassi risk estimate to get infection with *S. aureus*

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was (1.057) time compared with (0.986) for mixed breed, risk estimate for mixed breed to get infection with MRSA was (0.745) time.

C- Flock size:

As shown in table (4), *S. aureus* and MRSA infections were concentrated among small flock size 34/75 rated (30.67%) versus 11/75, (14.67%), also infection with *S. aureus* and MRSA reported among large flock size, 12/75, (16%) versus 3/75, (4%). Neither significant difference ($\chi 2$=0.216; p value =0.642), non-correlation (R= -0.054; p value =0.647) reported between flock size and *S. aureus* infections among sheep. Also Neither significant difference ($\chi 2$=0.369; P value = 0.544) non-correlation (R= - 0.070; P-value = 0.550) reported in the middle of flock size via MRSA infections among sheep. Probability of flock size (small/ large), for infection with *S. aureus* or (odds ratio) was 0.784 time compared with (0.652) for MRSA.Risk estimate for small size flock to get infection with *S. aureus* was (0.933) time compared with (1.190) for large size flock, risk estimate for small size flock to infect with MRSA was (0.897) time compared (1.377) for large size flock.

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Table (4): Flock Size As A Risk Factor For Infection With *S. aureus* and MRSA among sheep

Flock size	Type Of Isolates From Skin Lesions Of Sheep				
	S. aureus		MRSA		
	Positive	Negative	Positive	Nega-tive	Total
Small	34(30.67%)	20(26.67%)	11(14.67%)	43(57.33%)	54(72%)
Large	12(16%)	9(12%)	3(4%)	18(24%)	21(28%)
Total	46(61.33%)	29(38.67)	14(18.67%)	61(81.33%)	75(100%)
χ2	0.216		0.369		
P value	0.642		0.544		
R	-0.054		-0.070		
P value	0.647		0.550		
Odds Ratio for size	Value	95% CI	Value	95% CI	
flock(small/ large)	0.784	0.281-2.187	0.652	0.162 - 2.616	
Risk estimate for small size flock	0.933	0.692-1.258	0.897	0.653- 1.233	
Risk estimate for large size flock	1.190	0.574-2.465	1.377	0.470- 4.036	

D-Season:

As shown in table (5), *S. aureus* and MRSA infections were con-centrated in autumn 35/75, (46.67%) versus 10/75, (10.67%), although infection with *S. aureus* and MRSA reported in winter, 11/75, (14.66%) versus 6/75, (8%),Neither significant difference ($\chi 2=0.461$; p value =0.497), nor correlation (R= 0.078; p-value =0.504) reported amongst Season via *S. aureus* infections in sheep,

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Neither significant difference ($\chi 2$=2.307; p value = 0.129) non-correlation (R= - 0.175; p-value = 0.132) reported amongst Season via MRSA infections in sheep.

Table (5):Season as infection risk factor with *S. aureus* and MRSA in sheep

Season	Type Of Isolates From Skin Lesions Of Sheep				
	S. aureus		MRSA		
	Positive	Negative	Positive	Nega-tive	Total
Winter	11(14.66%)	9(12%)	6(8%)	14(18.67%)	20(26.67%)
Autumn	35(46.67%)	20(26.67%)	8(10.67%)	47(62.67%)	55(73.33%)
Total	46(61.33%)	29(38.67)	14(18.67%)	61(81.33%)	75(100%)
χ^2	0.461		2.307		
P value	0.497		0.129		
R	0.078		-0.175		
P value	0.504		0.132		
Odds Ratio for Season (Winter / Autumn)	Value	95% CI	Value	95% CI	
	1.432	0.507 - 4.044	0.397	0.118- 1.339	
Risk estimate for winter	1.298	0.614-2.743	0.536	0.250- 1.145	
Risk estimate for Autumn	0.906	0.676-1.215	1.348	0.839- 2.166	

The probability for infection with *S. aureus* in certain Season (winter / autumn), or (odds ratio) was 1.432time compared with (0.397) for MRSA, risk estimate to get infection with *S. aureus* in winter was (1.298) time compared with (0.906) for autumn, risk estimate to get infection with MRSA in winter was (0.536)

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time compared with (1.348) for autumn.

E-Introducing New Sheep to the Flock:

As table (6) shown, *S. aureus* infections reported in 16/75, (21.33%) of sheep wherever new sheep were introduce in to the flock versus 30 /75, (40%) of infection that reported without any renew for the original flock. Although, infection with MRSA was reported in, 3/75, (4%) of sheep, wherever new sheep introduce in to the flock versus 11/75, (14.66%) of infection that reported without any renew for the original flock, Neither significant difference ($\chi 2$=2.715; p value =0.099), non-correlation (R= -0.190; p-value =0.102) reported flanked by introducing new sheep to the flock and *S. aureus* infections among sheep. Neither significant difference ($\chi 2$=0.369; P value = 0.544), non-correlation (R= -0.070; p-value = 0.550) reported amid introducing new sheep to the flock and MRSA infections among sheep, probability for infection with *S. aureus* due to introducing of new sheep to the flock or (odds ratio) was 0.391 time compared (1.535) for MRSA. Risk estimate to get infection with *S. aureus* due to introducing of new sheep to the flock was (0.496) time compared (1.269) for infections without renewing of the original flock. On other hand, risk estimate to get infection with *MRSA* due to introducing of new sheep to the flock was (1.377) time compared (0.897) for infections without renewing original flock.

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Table (6): Renewing sheep flock as risk factor for *S. aureus*, MRSA infections

Introducing of new sheep to the flock	Type Of Isolates From Skin Lesions Of Sheep				
	S. aureus		MRSA		
	Positive	Negative	Positive	Nega-tive	Total
Yes	16(21.33%)	5(6.66%)	3(4%)	18(24%)	21(28%)
No	30(40%)	24(32%)	11(14.66%)	43(57.33%)	54(72%)
Total	46(61.33%)	29(38.67)	14(18.67%)	61(81.33%)	75(100%)
χ2	2.715		0.369		
P value	0.099		0.544		
R	-0.190		0.070		
P value	0.102		0.550		
Odds Ratio for Introducing of new sheep to the flock (yes/ no)	Value 0.391	95% CI 0.125-1.220	Value 1.535	95% CI 0.382- 6.163	
Risk estimate for Introducing of new sheep to the flock =yes	0.496	0.204-1.207	1.377	0.470- 4.036	
Risk estimate for Introducing of new sheep to the flock =NO	1.269	0.970-1.660	0.897	0.653- 1.233	

F-Wounds:

As shown in table (7), sheep wounds suffered 24/75, (32%), *S. aureus* was isolated from wounds 14/75, (18.67%). MRSA recovered from 6/75, (8%) of wounds cases too, probability for infection with *S. aure-*

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us due to wounds or (odds ratio) was 0.831time, the risk estimate to get infection with *S. aureus* due to wounds was (1.133) time compared with (0.942) intact skin the risk estimate to get infection with *MRSA* for sheep suffered from wounds was (0.689) time compared with (1.234) for infection that reported among sheep with intact skin.

Table (7):Sheep wounds as a risk factor for infection with *S. aureus*, MRSA

Type Of Skin Lesion	Type Of Isolates From Skin Lesions Of Sheep				
Wounds	*S. aureus*		MRSA		
	Positive	Negative	Positive	Negative	Total
No	32(42.67%)	19(25.33%)	8((10.67%)	43(57.33%)	51(68%)
Yes	14(18.67%)	10(13.33%)	6(8%)	18(24%)	24(32%)
Total	46(61.33%)	29(38.67)	14(18.67%)	61(81.33%)	75(100%)
Odds ratio for wound (No / yes)	Value 0.831	95% CI 0.309-2.238	Value 1.792	95% CI 0.543- 5.907	
Risk estimate for wound =yes	1.133	0.583-2.204	0.689	0.336-1.412	
Risk estimate for wound =No	0.942	0.680-1.305	1.234	0.762- 1.997	

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G- **Abscess:**

As shown in table (8), 22/75, (29.33%) sheep were suffered from Abscess *S. aureus* was isolated from 20/75, (26.67%) with Abscess. MRSA was recovered from 5/75, (6.67%) of abscess cases, the probability for infection with *S. aureus* due to Abscess or (odds ratio) was 10.385 time. The risk estimate to get infection with *S. aureus* due to Abscess was (0.159) time compared with (1.647) intact skin, the risk estimate to get infection with *MRSA* for sheep suffered from abscess was (0.780) time compared with (1.122) for infection that reported among sheep with intact skin.

Table (8):Sheep abscess as infection risk factor with *S. aureus* and MRSA.

Type Of Skin Lesion	Type Of Isolates From Skin Lesions Of Sheep				
Abscess	*S. aureus*		MRSA		
	Positive	Negative	Positive	Negative	Total
No	26(34.67%)	27(36%)	9(12%)	44(58.67%)	53(70.67%)
Yes	20(26.67%)	2(2.67%)	5(6.67%)	17(22.67%)	22(29.33%)
Total	46(61.33%)	29(38.67)	14(18.67%)	61(81.33%)	75(100%)
Odds ratio for Abscess (No / yes)	Value	95% CI	Value	95% CI	
	10.385	2.204-48.932	1.438	0.421- 4.910	

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Risk estimate for Abscess =yes	**0.159**	**0.040-0.629**	**0.780**	**0.347- 1.755**
Risk estimate for Abscess =No	**1.647**	**1.255-2.162**	**1.122**	**0.737- 1.708**

H- Dermatitis:

As shown in table (9), 17 /75, (22.67%) sheep were suffered from dermatitis *S. aureus* was isolated from 11/75, (14.67%) with dermatitis. MRSA was recovered from 2/75, (2.67%) of dermatitis cases. The probability for infection with *S. aureus* due to dermatitis (No / yes) or (odds ratio) was 1.205 time compared with (0.511) for MRSA, risk estimate to get infection with *S. aureus* due to dermatitis was (0.865) time compared with (1.042) healthy skin, while risk estimate to get infection with MRSA for sheep suffered from dermatitis was (1.721) time compared with (0.880) for infection that reported among sheep with intact skin.

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Table (9);Sheep Dermatitis as a risk factor for infection with S. aureus and MRSA

Type Of Skin Lesion	Type Of Isolates From Skin Lesions Of Sheep				
Dermatitis	*S. aureus*		MRSA		
	Positive	Negative	Positive	Nega-tive	Total
No	35(46.67%)	23(30.67%)	12(16%)	46(61.33%)	58(77.33%)
Yes	11(14.67%)	6(8%)	2(2.67%)	15(20%)	17(22.67%)
Total	46(61.33%)	29(38.67)	14(18.67%)	61(81.33%)	75(100%)
Odds ratio for Dermatitis (No / yes)	Value 1.205	95% CI 0.391-3.712	Value 0.511	95% CI 0.103- 2.548	
Risk estimate for Dermatitis =yes	0.865	0.359-2.085	1.721	0.443- 6.682	
Risk estimate for Dermatitis =No	1.042	0.815-1.334	0.880	0.680- 1.138	

I- Abrasion: Ass shown in table **(10),** 12 /75, (16%) sheep were suffered from abrasions, *S. aureus* was isolated from 1 /75, (1.33%) with abrasions. MRSA was recovered from 1/75, (1.33%) of abrasions cases, the probability for infection with S. *aureus* due to abra-

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sions (No/ yes) or (odds ratio) was 0.036 time compared with (0.350) for MRSA, the risk estimate to get infection with S. aureus due to abrasions was (17.448) time compared with (0.634) healthy skin, the risk estimate to get infection with MRSA for sheep suffered from abrasions was (2.525) time compared with (0.883) for infection that reported among sheep with intact skin.

Table (10): Sheep Abrasion as a risk factor of infection with *S. aureus* and MRSA in

Type Of Skin Lesion	Type Of Isolates From Skin Lesions Of Sheep				
Abrasions	*S. aureus*		MRSA		
	Positive	Negative	Positive	Negative	Total
No	45(60%)	18(24%)	13(17.33%)	50(66.67%)	63(84%)
Yes	1(1.33%)	11(14.67%)	1(1.33%)	11(14.67%)	12(16%)
Total	46(61.33%)	29(38.67)	14(18.67%)	61(81.33%)	75(100%)
Odds ratio for Abrasions (No / yes)	Value 0.036	95% CI 0.004-0.303	Value 0.350	95% CI 0.041- 2.960	
Risk estimate for Abrasions =yes	17.448	2.376-128.112	2.525	0.355- 17.977	
Risk estimate for Abrasions =No	0.634	0.476-0.846	0.883	0.732- 1.064	

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Discussion:

Samples collected from Kanaan, Baladruze, Baqubah and Buhruz region, this area flat land, same soil contains, extension unity, no natural barriers except main Diyala river, no hills or mountains, represent same tail end of its rivers, no oil wells, refineries or big factories which might contaminate atmosphere with carbon mono or dioxides, no mine prospecting, and same rain fall ratios, located middle east extent to south part of Diyala Province.

Methods of breeding, housing and feeding domesticated animals vary from species to species and differ according to custom and accordance with geographical and climatic conditions**[**31**]**. Current study revealed that *S. aureus* rates of skin infection among sheep was 61.33%, rates of MRSA was 30.43 %, by means of traditional biological tests include; MSA which isolate, selects and differentiated; G positive cluster like coccid positive for Coagulase, Catalase DNase tests respectively according to **[**32**,** 33**]** then confirmed by Vitek2 system which goes with**[**34**,** 35**]** later conventional PCR by using *S. aureus* 23sRNA gene sequence specific primer (staur4,6)**[**36**,** 37**]** . whereas MRSA was 14/46, rated (30.43%) among positive isolates representing (18.67%) from total samples 14/75 according to methicillin resistance via Muller Hinton medium depending on **[**38**,** 39**]** placed by results of conventional PCR using *S. aureus mec*A gene **[**5**,** 40**]**.

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Infection rates varied around the world, while In north-western Greece[41] found that out of 367 samples tested, 57.8% were *S. aureus* and only 3% MRSA positive, although in Bangladesh the rate was higher for *S. aureus*, 70% [31]. In Italy [42] recorded that infection rate of *S. aureus* among dairy sheep farms was 53.5% and 7% for MRSA among the hall flock. French farms study showed nasal carriage of *S. aureus* in 29% of dairy ewes[43] . In Norway [44] recoded *S. aureus* was (32.6%) in sheep. In Morocco similar study [45] revels low in sheep (9.97%) These variations might be due to differences in sample size, isolation techniques, awareness and skills of the farm workers, geographic regions and variation in study subjects, the different in management system used by the farm, types of sample, diagnostic test.

Sex factor recorded *S. aureus* infections among sheep female was in (41.33%), MRSA (12%), non-significant higher than male (20%), MRSA (6.66%), thought to be exposing to heavily impacts; manually milk lactation twice a day for the breeder's benefits, extended udders, suckling lambs, periodic mastitis, parturitions or recurrent abortions, raises the infection ratio, which goes with[7] whom explains correlation between *S. aureus*, MRSA and sheep sex, also [7] assumed that other factors associated with higher incidences include males which is unlikely to be a major factor. Infections among males were higher, it might be regarded to the source of collected samples[46]. In current study although there was no significant correlation between sheep sex and infection with *S. aureus* and MRSA, the possible explanation for these results may attribute to the role of sex hormones in modulation of immune response and susceptibility for infection among males and fe-

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males. Steroid hormones have important role in regulates skin physiology and immunity, skin architecture, thickness of dermis and epidermis layers[47, 48] Estrogens associated immune-enhancement while androgens with immunosuppression[7, 49] While Estrogens play a protective role against S. aureus derma necrosis. On the other hand, female-biased transcriptomic signature in the skin that is independent of sex steroid levels have the possibility for increase the rate of infection among females[50] .

In current study, no correlation was reported between breed and *S. aureus* infections among sheep. Significant correlation was reported between breed and MRSA infections, probability of breed (Awassi / Mixed), to infect with *S. aureus* was (1.072) time, risk estimate for Awassi to infect by *S. aureus* was (1.057) time compared with (0.986) for mixed breed, Risk estimate for mixed breed to infect with MRSA was (0.745) time, these results may have attributed to the popularity of Awassi among submitted flocks, in Arabia Saudi [46]reports Najdi breed infected rates (22.4 %) followed by the Sakni breed (20%) then Naimi breed (4.8 %), he pointed that some sheep breeds were found to be more susceptible to the disease than others, also in Italy [42] report Marino breed being highest, [51] claimed that race effects on resistant to infection was genetically.

No significant correlation was reported between flock size and *S. aureus*, MRSA infections among sheep. The probability of small flock size versus large flock size for infection with *S. aureus* was (0.784) time compared with (0.652) for MRSA.

In Italy [6], claimed that emergence of livestock associated MRSA correlate with farm size, and animal trading spread LA-MRSA inside, she

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concludes that picture of MRSA transmission among sheep farmers doesn't seem critical, she recommended to improve adequate control measures to prevent and minimize any biological risk for both human and animal health, despite her suggests, results might tell more effect factors about unlashed *S. aurues* and MRSA high rated infections. Likewise, flock irregular management, external parasite treatments, dipping, chemical spraying, and wool shearing; also *S. aureus* affected 53.33% in semi-intensive farm animal and 66.67% in intensive farm **[**31**]** also**[**52**]** pointed that LA-MRSA was higher in larger farms than in smaller farms (34% versus 7%, respectively). Thus **[**53**]** propose that risk of outbreak correlate and increased with larger flock size especially flocks with a lambing percentage greater than 200.

Season risk estimation to get infection with *S. aureus* in winter was higher compared with autumn, while risk estimation to get infection with MRSA in winter was lesser compared with autumn, that might be parallel to early birth season, lamb feeding, decreasing in temperature, weather changing, hours of a day lighting, starting new agricultural cycle seasons to planting by the end of summer season where is green leafs turned to dryness. Although **[**54**]**resume spring and autumn were the most popular seasons for CA-MRSA isolates, while HA-MRSA infection often occurred in summer and winter, hence **[**53**]** give a warning to Norwegian sheep flocks farmers about winter housing risks of Staphylococcal infectious arthritis.

The risk estimation to get infection with *S. aureus* due to Introducing new sheep to flock was (0.496) time compared with out renewing of the original flock (1.269) time, (Mascaro, et al.2019) in Italy claimed that emergence of LA-MRSA increase through

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animal trading which spread LA-MRSA inside, also [55]in Sudan confirmed that.

Wound risk estimation to get infection with *S. aureus* due to wounds was (1.133) time compare with intact skin was (0.942) time, while risk estimation to get infection with MRSA higher. On the other hand [31] explain that all kinds of wounds on skin and hides usually occur due to breach with very sharp instruments, imperfect brand marks, rubbing against course surfaces or incisions made by doctors during surgical operations, get contaminated by a big population of bacterial flora such as pyogenic bacteria especially *S. aureus*, *Streptococcus pyogenes*, and Coliform group together with *Proteus vulgaris welchii* type A, *Clostridium septicum*, *Clostridium diphtheriae*, cause damage to the qualities of leather which reflect in sense of losses to the leather industries, economy in addition being life threating. Young lambs early ear tagging associated with an increased risk of outbreak due to ear wounds increase the infection rats 5.6% [53].

Wounds is circumstances sporadic event, needs sharp object to induce wounds later contamination take a place, but massive occurrences related to season of wool shear, sexual meeting season where rams get horn fights also spring - summer period companied with external parasites (ticks, mosquitos, screw warms, lice and mites), Surgical operation, needles penetration, dehorning.

Abscess led to huge economic losses in sheep industry during Haj season[46] with high mortalities. prevalence of abscess disease varied between 5%– 44.1% in sheep farms, highest in the Najdi breed, Abscess forming 25.77 % were due to *S. aureus* disease which is a worldwide contagious disease of sheep, adversely affecting the development of the

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sheep industry known as Morel's, disease and caseous lymphadenitis, contagious nature, worldwide distribution, occurred around the time of shearing. On the other hand , [55] stated that many Sudanese shipments of export sheep were rejected by Saudi Arabia on the grounds of this disease, economic losses of each amounted millions of US dollars. In current study, abscess with *S. aureus* infection rated (26.67%), MRSA (6.67%), in Sudan [55] claimed normally encountered in lambs between 4 and 10 months of age and is characterized by the formation of abscesses in or adjacent to superficial lymph nodes, *S. aureus* was isolated in pure cultures samples (43.79%), In Arabia Saudi[46] reports infection rates (12.37 %) among infected sheep with abscess.

Dermatitis risk estimation to get infection with *S. aureus* was (0.865) time compare with healthy skin was (1.042) time, while risk estimation to get infection with MRSA due to Dermatitis was (1.721) time compare with healthy skin was (0.880) time. On the other hand [56]described dermatitis as a primary disease caused by *S. aureus* in sheep skin, also causes acute gangrenous mastitis before and after parturition which is highly persistent during lactation in ewes and lambs that nurse on milk or colostrum from a doe with acute mastitis can develop staphylococcal dermatitis, ovine facial staphylococcal dermatitis is mainly localized around the ocular area of adult sheep and tends to be seasonal, Sucking flies seem to be a potential seasonal factor of facial staphylococcal dermatitis, expanding the lesions and spreading diseases within a flock. An epidemiological study revealed that more than 50% were *S. aureus* positive during the summer season, which can turned to ovine necrotic dermatitis, that often

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occurs on the legs and above the lips, with potential advancing to be another skin disease called contagious ovine digital dermatitis. Dermatitis with *S. aureus* infection rated (14.67%), MRSA (2.67%), thought that even internal parasites could play a role, in Norwegian Veterinary Institute. On the other hand **[44]**recorded *S. aureus* samples from Dermatitis body swab (32.6%) despite lambs (58.2%).

Abrasions risk estimation to get infection with *S. aureus* due to abrasions was (17.448) time far away to compare with healthy skin which was (0.634) time, while risk estimation to get infection with MRSA due to Abrasions was (2.525) time compare with healthy skin was (0.883) time, **[57]** explained that *S. aureus* dermatitis typically involves trauma previously due to the close contact of heads over feeding troughs and abrasive plants at pasture may be predisposing factors, although its riskiness, multiple exposures might thickened the skin area, alarmed the immune system, and twist the wool strata.

While abrasions *S. aureus* infection rated (1.33%), MRSA (1.33%). that goes with**[58]** mention increasing risk factors were skin abrasions or wounds, contacts, overweight or obesity, antibiotic use within 6 months, overcrowding during feeding or housing with another animals like cattle.

Conclusions:

Ewes have greater risk to get infection with *S. aureus* and MRSA .

Significant correlation was reported between infection with MRSA and breed of sheep. Awassi have greater risk for infection with *S. aureus* **.** No significant correlation was reported between the flock sizes, the season, introducing of new sheep to the flock, wounds, dermatitis and *S. aureus*, MRSA infections among sheep, while Sheep S.aureus skin infection corre-

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lated significantly with abscess and abrasions.

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