STAPHYLOCOCCUS AUREUS OF CANINE NOSTRIL ORIGIN: BACTERIOPHAGE TYPING, ANTIBIOTIC SENSITIVITY, AND BIOCHEMICAL CHARACTERISTICS OF ISOLATED CULTURES

by

HAROLD EDWARD GARNER

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Approved by:

Major Professor

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INTRODUCTION

Staphylococcic infections in man have become increasingly important in the past several years and are of particular significance when antibiotic resistant strains develop. The source of the infecting organism in man has become of paramount importance in attempting to control this condition. Results of surveys conducted on limited numbers of animals suggest that they might serve as a reservoir for such human infections. Since the dog is intimately associated with man, a study of the carrier state of this animal might reveal one of the sources of human infections. Therefore, an attempt was made in this study to substantiate an interspecies relationship of pathogenic staphylococci. Such an interspecies relationship is probably most adequately demonstrated by the utilization of bacteriophage typing accompanied by antibiotic sensitivity determinations.

One of the constant characteristics of any strain of Staphylococcus aureus is its susceptibility to bacteriophage. However, very little information is available comparing the various measures of pathogenicity of staphylococci of canine origin to bacteriophage typing, and consequently this was incorporated as a part of the study.

REVIEW OF LITERATURE

Distinguishing Principles of Staphylococci. Many methods of determining the pathogenicity of staphylococci have been described; however, only a few of these methods have been found

to be true measures of the organisms' pathogenicity. Early workers screened the staphylococci via mannitol fermentation, gelatinase production, hemolysis of erythrocytes, and colony pigmentation with somewhat limited success. More recently, the coagulase test and bacteriophage typing have been developed and used successfully for identifying staphylococcic pathogens.

In 1934 Chapman, in his study of 5,000 strains of Staphylococcus aureus, found that coagulating strains were usually pathogenic regardless of hemolytic activity or pigment production. Cruickshank in 1937 considered the presence of staphylocoagulase to be a characteristic only of the pathogenic members of the species whether aureus or albus. It appeared to be an enzymic substance acting in a manner similar to thrombin upon the plasma of various animal species, including man. Christie and Keogh (1940), Fairbrother (1940), Bell (1940), Field and Smith (1945) all substantiated the coagulase test as the only reliable determination of pathogenicity for Staphylococcus aureus.

Chapman (1934) found that coagulase activity and hemolysis were usually parallel reactions; however, instances occurred in which one was strongly positive and the other entirely negative. Minett (1936) said the production of beta toxin was a characteristic feature of hemolytic staphylococci from animals. Both Smith (1947) and Minett found that dog strains produced small or undetectable amounts of alpha toxin. Bell (1940) found no correlation between alpha and beta toxin production, presence of coagulase, mannitol fermentation, or gelatinase production.

Mannitol fermentation, gelatinase production, and pigment production have been found by the previously mentioned workers and others to be inconsistent properties and unreliable measures of pathogenicity of staphylococci. Smith (1947) stated that the possession of any one of these properties may be accepted as evidence of pathogenicity, but its absence was without significance.

Antibiotic Sensitivity of Staphylococci. The increased use of antibiotics resulted in the development of resistant strains of staphylococci. The data obtained by Live and Nichols (1961) indicated that antibiotic resistant strains of staphylococci became established in veterinary hospitals, and these hospitals were a source of human infection and infection of hospitalized animals.

Williams et al. (1953) found staphylococcic strains in bacteriophage group III to be, in most cases, more penicillin resistant and to be more common in the feces than in the nose of humans. In a milk cow herd study, Price (1954) found that phage type and penicillin sensitivity were stable for several months in animals infected with staphylococci.

Later, Blair and Carr (1958) found resistant strains isolated from man to be usually resistant to penicillin and streptomycin and 10% of the strains to be resistant to the tetracyclines, erythromycin or chloramphenicol. All strains were resistant to polymyxin. All strains were sensitive to bacitracin, carbomycin, neomycin, and novobiocin. Group I

(29, 52, 52A, 79) led other groups in resistence with 16% to 33% resistant to penicillin, streptomycin and the tetracyclines.

Zinn (1961) examined samples of milk, swabs from human nasal cavities and hands, cattle lesions, and machine inflations which revealed the repeated presence of staphylococcic strains resistant to penicillin, streptomycin and chlorotetracycline. Mann (1960) tested 31 cultures isolated from dogs and cats of which 18 were sensitive to penicillin, 25 to bacitracin, 29 to oleandomycin-tetracycline, and all to oleandomycin.

Bacteriophage Typing. The result of the development of routine staphylococcic screening tests was the adaptation and acceptance of these methods by most of the laboratories dealing with the staphylococcic problem. However, with the ultimate and simultaneous development of antibiotics and sulfa drugs, new problems arose, and thus another measure for screening staphylococci developed, namely, bacteriophage typing. The first phage typing laboratory was set up in the Central Public Health Laboratory and Colindale, London, England under Dr. V. D. Allison and Dr. R. E. O. Williams in 1946.

Twort in 1915 and d'Herelle in 1917 (Rejula, 1959) observed bacterial lysis by a transmissible lytic agent. Bacteriophage, or "phage" was the term applied to this lytic, ultramicroscopic and filterable agent. D'Herelle developed a quantitative approach using both lysis in broth and the production of described clearings or plaques on confluent surface

bacterial growth on agar (Burnett, 1955).

Sensitivity to bacteriophage has been the criteria used in the classification and differentiation of individual strains of several bacterial species. The phages have become valuable tools for the taxonomic and epidemiological studies of bacterial infections due to their selective action against strains of bacteria within the same species. The susceptibility of particular strains of an organism to phage action and the specific action of the phage are constant and stable characteristics (Blair, 1956).

Anderson and Williams (1956) described a "type determining phage" as a temperate phage, the presence of which in the cell exercises a partial control over the sensitivity of a bacterial phage type to a range of typing phages.

Craigie and Yen (1938) described a method of demonstrating types of Salmonella typhosus by means of preparation of type II Vi phage. They found type II Vi phage exhibited a high selective affinity, not only for particular V strain of S. typhosus on which it was propagated, but also epidemiologically related strains. On this basis a number of distinct phage types of S. typhosus have been recognized. Since then the techniques for propagation of the phages and the actual phage typing have been simplified.

Fisk (1942) found bacteriophage carriers among normal strains of Staphylococcus aureus not to be uncommon. He found the incidence of lysis to be 44.2% in his study. Lysogens

could not be demonstrated among cultures lacking characteristics of pathogenic S. aureus, and these phages exhibited selective activity when tested on strains among the aureus group. It was by this manner that he identified 24 different phages. He was unable to obtain lysis in broth.

Wilson (1945) succeeded in typing 60.4% of 1,340 strains of staphylococci and found organisms within Group I were from fulminating cases of pneumonia usually complicating influenza. Food poisoning strains were classified in Group III. Strains isolated from boils, otitis, and whitlows were shown often to have the same type as those strains found on normal individuals.

Blair and Carr (1953) determined the sensitivity of 539 coagulase-positive strains of staphylococci. Of these, 290 (53.6%) were susceptible to one or more of a series of 25 phages and were assigned to phage types, 4 (0.7%) gave equivocal results and could not be assigned to type, and 245 (44.7%) were not lysed by any phage of the series.

Later, Blair and Carr (1958) found the phage types usually encountered in the majority of antibiotic resistant staphylococci to be 80/81. Next in order of frequency were Groups I, II, and IV. They claimed that 80% of the antibiotic resistant strains were of Group III (6, 7, 42E, 47, 53, 54, 70, 73, 42B, VA4, 74, 75). They found 52A alone, 52A/79, and 52/42B/47C/44A were all incriminated in individual outbreaks. Also 53/77 and VA4 were found responsible for hospital outbreaks. They observed outbreaks of the 80/81 strains to be almost explosive,

being serious clinically in the form of neonatal infections, breast abcesses, furuncles, carbuncles, pneumonias, and surgical wound infections.

Levy (1953) found that with only one exception, the animal strains were within Group III or were unclassifiable or untypable. The untypable and Group III strains seemed to produce very little pigment, less fibrinolysin, and more beta hemolysin when compared to the other groups.

Smith (1948) successfully typed 93.3% of 1,016 staphylococcic strains isolated from milk samples. He found that it was usual to find one predominant phage type within one herd of cows. Although the phage typing did not distinguish mastitis udders from normal udders, it was found to be useful in the study of the epidemiology of bovine mastitis. Three strains, one food, one human mastitis, and one bovine mastitis were typed by 42D, were antigenicly identical, and maintained themselves in bovine udders for six weeks. He also found that bovine strains were susceptable to another set of phages.

Price *et al.* (1954), in a three herd study dealing with penicillin sensitivity and phage typing, discovered that two phage types were rarely found in established infections within one quarter, even when there were three or four phage types within the herd.

Wallace (1960) reported three cows in one dairy to have mammary glands infected with the antibiotic resistant staphylococcic phage type 80/81. Subsequent investigations revealed

one dairy employee and his family to be suffering from staphylococcic disease caused by the same type. This organism was found in 20 to 21 raw bulk milk samples. Treatment on one of the cows failed to halt the excretion of 80/81 in the milk.

Likewise, Zinn et al. (1961) revealed repeated presence of staphylococcic phage type 80/81 in cultures of milk, human nasal cavities and hands, cattle lesions, and machine inflations. Their epidemiological evidence indicated that staphylococci were not always species specific. They reported furunculosis in dairy cows as being a problem associated with dairy attendants who were undergoing treatment for sinusitis and otitis.

From the nasal swabs of a number of domestic and laboratory animals, Rountree et al. (1956) found the majority of the staphylococci isolated to be similar in phage type to those isolated from man. Seven staphylococcic carriers were found among 35 hospitalized dogs, while no carriers were found in 26 "out patient" dogs. Phage patterns found in dogs by Rountree were as follows:

7/42E/47	2
7/47	1
31B/44	2
42E/47	1
No Type	3

Mann (1959) cultured the nasal swabs from 100 dogs and 100 cats. Two of the cats yielded phage-typable strains of Staphylococcus aureus similar to those infective for man.

In 1960 he phage typed 228 animal isolates and found strains similar to phage types of S. aureus that infected man. He typed three bovine strains with 80/81 and obtained the following results with those isolated from dogs:

3A	1
7A	1
29/52/80	1
42D/52A/71/80	1
52/80/81	1
53/75/77/83	1
187	1
No Type	23

Live and Nichols (1961) found 4.9% of 445 out patient dogs carried in their anterior nares staphylococci typable with the international series of phages. Only 2 of the cultures (0.4%) were phage type 80/81, and a large majority of all the isolates were sensitive to the antibiotics used.

Coles (1959) adapted human typing phages to animal strains of staphylococci indicating the possibility of adapting additional phages for routine typing of animal cultures.

Rajula (1960) isolated 28 coagulase-positive strains from 30 dogs. The number producing hemolysis correlated closely to coagulase production, while mannitol fermentation, gelatinase production, and aureus pigmentation seemed to be less significant. He found canine strains typable with international phages, those typed by Group III being more resistant

to antibiotics than the other groups.

Pagano et al. (1960) phage typed the staphylococcic strains carried by the animals and senior students in a veterinary hospital and found phage type 80/81 predominated.

Fratta (1960) phage typed 30 of 66 staphylococci isolates of canine origin with the international phages at RTD and typed 39 at 100 X RTD. They suggested the use of both dilutions in typing animal strains.

MATERIALS AND METHODS

The nasal cavities of dogs were examined for the presence of staphylococci. Many (335) of the dogs included in this study were patients at the Dykstra Veterinary Hospital, 29 were owned by the Kansas State University Pathology Department, and 18 were from the Sea Bee Boarding Kennels of Manhattan, Kansas.

Mannitol Fermentation. The external nares of each dog was swabbed with a sterile cotton swab that was placed in a sterile stoppered tube of trypticase soy broth* and incubated at 37 C. overnight. These swabs were then seeded on mannitol salt agar for isolation of staphylococci. Phenol red was an ingredient of this broth to indicate the fermentation of mannitol. A change from red to yellow within 48 hours indicated mannitol fermentation.

Hemolysis. After isolation on mannitol, the staphylococci were transferred with a sterile loop to 5% sheep blood

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*Baltimore Biological Laboratories, Baltimore, Md.

agar. They were incubated 24 hours at 37 C. and observed for the production of alpha and/or beta hemolysis.

From the blood agar the colonies were transferred to agar slants for storage and further study.

Coagulase Production. The coagulase test was performed on all isolated staphylococci from 12 hour broth cultures, using rabbit plasma. To 0.5 ml. of plasma was added two drops of the broth culture. These tubes were then incubated for 48 hours at 37 C. Readings were made at one-half hour, 1, 3, 24, 36, and 48 hours. Any clotting during this period of time was considered positive. All strains were screened for purity by the Gram stain.

Pigment Production and Gelatinase Production. All coagulase positive staphylococci were grown on Chapman Stone Agar* to determine pigment production. Aureus, citreus, and albus were the determinators used. The colonies on these same plates were flooded with 5 ml. of a saturated solution of ammonium sulfate and allowed to stand for 10 minutes. Clear zones around the colonies indicated a positive gelatinase reaction.

Antibiotic Sensitivity. Antibiotic sensitivity was determined on all of the coagulase positive strains. The organisms were swabbed on tryptic soy agar* from 12-hour broth cultures. When dry, the plates were spotted with antibiotic discs** and incubated for 12 hours at 37 C. The

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*Difco Laboratories, Detroit, Mich.
**Case Laboratories Inc., Chicago 22, Ill.

discs contained the following concentration of antibiotics: chlortetracycline, 5 g.; tetracycline, 5 g.; oxytetracycline, 5 g.; erythromycin, 2 g.; neomycin sulfate, 5 g.; penicillin, 2 units; phenoxyethyl pencillin, 2 g.; sodium dimethoxyphenyl penicillin, 5 g.; chloramphenicol, 5 g.; dihydrostreptomycin, 2 g.; and bacitracin, 2 units. A clear zone of 10 mm. diameter around an antibiotic disc indicated positive sensitivity for the strain being used.

Bacteriophage Typing. The following bacteriophages of the international series were used:*

29, 52, 52A, 79, 80	Group I
3A, 3B, 3C, 55, 71	Group II
6, 7, 42E, 47, 53, 54, 75, 77	Group III
42D	Group IV
81, 187	Miscellaneous

Phages 83, 73, 44A, and the new strains of bacteriophage identified as C-1, C-2, and C-4 ("C" phages) also were used. The "C" phages were isolated from lysogenic strains of Staphylococcus aureus of canine origin by Coles (1963). Phages A10, A8 and A13 were phages adapted to bovine staphylococci, and P52 and P87 were each isolated from a lysogenic strain of S. aureus of bovine origin (Coles, 1959).

Phages S-1, S-2, S-3, and S-5 used in the phage typing of bovine staphylococci (Seto and Wilson, 1958), also were tested for their ability to lyse these isolates.

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*The bacteriophages and their propagating hosts were furnished by Dr. J. E. Blair, Hospital for Joint Diseases, New York, N. Y.

Bacteriophage propagation was carried out according to the recommendation outlined by the C.D.C.* Each bacteriophage was propagated by the freeze-thaw method in which 15 ml. of soft agar (0.5% agar), inoculated with the phage and its propagating host, was overlaid on an agar base (1.5% agar) in an 8-oz. prescription bottle. Cultures were incubated 4 hours at 37 C. and then overnight at room temperature. They were then frozen; liquid which accumulated after thawing was collected and centrifuged at 2,000 r.p.m. for 30 minutes. The supernatant fluid was filtered through a Selas 03 candle,** and the filtrate was collected in sterile tubes and stored at 4 C. Phage propagated in this manner was titrated to determine the RTD. A drop of each tenfold dilution was added to a petri dish containing trypticase soy agar*** that had been previously seeded with the propagating host. The RTD was considered to be that dilution which just failed to give complete lysis on its propagating strain.

One and one-half per cent trypticase soy agar was poured into sterile petri dishes and then dried in the incubator at 37 C. for 24 to 48 hours. Stock cultures of staphylococci to be typed were transferred to trypticase soy broth and incubated 12 hours at 37 C. Four hour old trypticase soy broth cultures transferred from broth subcultured on trypticase soy

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*Communicable Disease Center, Atlantic City, N. J.

**Selas Filter Corporation of America, Dresher, Penn.

***Baltimore Biological Laboratories, Baltimore, Md.

agar previously incubated 18 hours at 37 C. were inoculated onto dry trypticase agar plates with sterile cotton swabs and with the aid of turntables. The plates were then marked at the top and allowed to dry before being inoculated with phage. Phage inoculation was accomplished with the aid of a dispenser.* After the dropping of the phage and allowing the drops to dry, the plates were placed in the incubator at 37 C. for 4 hours and then removed and incubated at room temperature for 8 to 12 hours.

Both the RTD and 100 X RTD were run on each coagulase positive strain. The patterns were read using a bright light. The following symbols were used in recording the degree of lysis at RTD and 100 X RTD:

Confluent lysis	+++
50 plaques and over	++
20-50	+
Less than 20	+

Only +++ and ++ were considered significant and were recorded as a part of the phage type or pattern of the isolate.

All phages were titrated 24 hours before a group of cultures was to be tested, and fresh phage dilutions were prepared the same day the typing was completed. All of the propagating strains for the phages being used also were plated and tested for their susceptibility to the bacteriophages. If any lytic reaction occurred other than those

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*Accu-Drop phage dispenser, manufactured by Accu-Tech Corp., New York, N. Y.

described by Blair and Williams (1961), the bacteriophage giving the abnormal reaction was repropagated from the original phage stock and the cultures were retested for their susceptibility.

DISCUSSION AND RESULTS

Coagulase Production. Staphylococci were isolated from 343 (89.7%) of the 382 dogs examined. Two hundred fourteen (62.1%) of these dogs carried coagulase-positive strains of staphylococci. Coagulase production was observed on rabbit plasma only.

Mannitol Fermentation. One hundred thirty-seven (64.1%) of the coagulase-positive staphylococcic strains isolated fermented mannitol. The remaining 77 (35.9%) strains did not ferment mannitol (Table 1).

Pigment Production. Of the 214 coagulase-positive staphylococcic strains, 155 (77.1%) produced citreus type colony pigmentation, 22 (10.3%) aureus pigmentation, and 27 (12.6%) produced no pigmentation or the albus type of colony growth (Table 1).

Gelatinase Production. Sixty-one (28.5%) of the coagulase positive strains of staphylococci produced gelatinase, and 153 (71.5%) did not.

Hemolysis. One hundred fifty-four (72.0%) of the coagulase-positive strains (214) produced some type of hemolysis. Ninety-nine (46.3%) produced double zone hemolysis

TABLE 1 -- Comparison of biochemical characteristics and bacteriophage typeability of 214 coagulase positive isolates of Staphylococcus aureus from canine nostril.

Item	No. and % of isolates possessing characteristic	Characteristic									
		Mannitol	Gelatinase	Hemolysis			Pigmentation				Phage type.
				∝	β	DZ	au	ct	al		
Mannitol fermentation	137 64.1%	137 64.1%									
Gelatinase production	61 28.5%	24 11.2%	61 28.5%								
Hemolysis	154 72.0%			154 72.0%							
alpha	54 25.2%	30 14.0%	14 6.5%	54 25.2%							
beta	1 0.5%	1 0.5%	0 0%	1 0.5%							
double zone	99 46.3%	44 20.6%	23 10.7%	99 46.3%							
Pigmentation											
aureus	22 10.3%	22 10.3%	8 3.7%	6 2.8	1 0.5	9 4.2%	22 10.3%				
citreus	155 77.1%	73 34.1%	34 15.9%	40 18.6	0 0	68 31.8%		155 77.1%			
albus	27 12.6%	7 3.3%	5 2.3%	6 2.8	0 0	8 3.7%			27 12.6%		
Phage Typable											
RTD	29 13.5%	20 9.3%	5 2.3%	10 4.7	0 0	7 3.3%	11 5.2	9 4.2	2 0.9%	29 13.5%	
100 X RTD	43 20.1%	25 11.7%	5 2.3%	14 6.5	0 0	8 3.7%	12 5.6	18 8.4	6 2.8%	43 20.1%	

(both alpha and beta), 54 (25.2%) produced alpha hemolysis, and one (0.5%) produced beta hemolysis.

Comparison of Biochemical Properties. Seventy-seven (35.9% of coagulase-positive strains) that did not ferment mannitol produced hemolysis. Forty-four produced double zone hemolysis, 30 alpha hemolysis, and one beta hemolysis.

Only 13 of the coagulase-positive strains fermented mannitol and produced gelatinase, some type of hemolysis, and either aureus or citreus colony pigmentation. Only one of these strains was bacteriophage typed (80/81)(Table 1).

Of the biochemical properties possessed by the coagulase-positive strains of Staphylococcus aureus, hemolysis production, mannitol fermentation, and citreus pigmentation were the most closely related to coagulase production. Aureus pigmentation and gelatinase production correlated to a lesser degree and were therefore not significant as measures of pathogenicity. These results did not disprove previous findings that the coagulase test for pathogenicity is the most reliable, but indicated the comparative value of the other biochemical tests.

Antibiotic Sensitivity. The coagulase-positive strains (214) of staphylococci were sensitive to eleven antibiotics as recorded in percentages in Table 2. Four strains were resistant to both penicillin and phenoxyethyl penicillin, 13 to penicillin and dihydrostreptomycin, 12 to the three tetracyclines, five to the tetracyclines and dihydrostreptomycin, three to the penicillins and the tetracyclines, and 40 to three or more groups of antibiotics.

TABLE 2 -- Antibiotic sensitivity of 214 *Staphylococcus aureus* cultures of canine origin.

Antibiotic	% of cultures sensitive
Chlortetracycline	80
Tetracycline	80
Oxytetracycline	70
Erythromycin	90
Neomycin sulfate	90
Penicillin	75
Phenoxyethyl penicillin	70
Sodium dimethoxyphenyl penicillin	81
Chloramphenicol	95
Dihydrostreptomycin	60
Bacitracin	87

The canine nostril strains in this study were more resistant to dihydrostreptomycin (40%), phenoxyethyl penicillin (30%), and oxytetracycline (30%) than to the other antibiotics used. In comparison Coles' (1963) isolates from other areas of the dog were more resistant to dihydrostreptomycin, penicillin, and oxytetracycline. Isolates from canine mouth and pharynx in Mann's (1960) study were more resistant to dihydrostreptomycin and tetracycline.

Apparently, the canine nostril strains in this study were more resistant to every antibiotic used than were the strains isolated by Coles (1963). Further study would be necessary to

elucidate the relationship between location of the staphylococci on the animal and pathogenicity.

Comparison of Antibiotic Sensitivity to Bacteriophage Typing. Eleven of 18 strains typed at RTD by the international series of bacteriophages were resistant to antibiotics. Seven of 10 strains typed by the "C" phages were resistant to antibiotics. Of the 43 strains phage typed at 100 X RTD, 20 were resistant to one or a combination of antibiotics (Table 2). The strains typed at RTD correlated more closely to antibiotic resistance than did those typed at 100 X RTD.

Bacteriophage Typing. Of the 214 strains exposed to bacteriophages, 43 (20.1%) were typed at 100 X RTD and 29 (13.5%) at RTD. The "C" phages were more active than those of the international series. Ten of the 29 isolates phage typed at RTD were typed by "C" phages, 21 by various combinations of the international series (3 were typed in combination with the bovine phages), and one by S-5. Seventeen of the 43 isolates phage typed at 100 X RTD were typed by "C" phages, 23 by the international series (3 in combination with bovine phages), and one by S-5 (Table 3).

The findings in this study indicated that some dogs, though only a small per cent (13.5%), do harbor strains of staphylococci identical to some of those harbored by man. This suggests the dog could be a carrier for those strains he has in common with man. Furthermore, it is possible that some of the many untypable staphylococci strains of canine

TABLE 3 -- Phage types among isolates of Staphylococcus aureus isolated from canine nostril at RTD and 100 X RTD.

No. Cultures	No. typed	Phage Dilution	Number of Cultures by Phage Types													
			Human Group						C1 C2	C4	80 81	80 81 52A	80 52A	80 81 52 52A	S-5	NT
			I	II	III	IV	MSC.MIX									
214	29	RTD	1	0	8	0	2	2	6	4	1	1	1	2	1	185
214	43	100 X RTD	2	0	8	0	0	8	12	5	1	0	1	3	1	171

nostril origin also are present in man. Additional studies need to be made to determine the public health significance of the presence of Staphylococcus aureus in dogs.

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STAPHYLOCOCCUS AUREUS OF CANINE NOSTRIL ORIGIN: BACTERIOPHAGE TYPING, ANTIBIOTIC SENSITIVITY, AND BIOCHEMICAL CHARACTERISTICS OF ISOLATED CULTURES

by

HAROLD EDWARD GARNER

B. S., Kansas State University, 1957
D.V.M.,Kansas State University, 1962

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This study was an attempt to reveal evidence concerning the interspecies relationship of staphylococcal infections between man and dog.

Two hundred fourteen coagulase-positive strains of Staphylococcus aureus were isolated from the external nares of dogs. Since bacteriophage typing and antibiotic sensitivity have seemed to be the most significant measure of pathogenicity in previous studies, they were employed in this study. Because there has been very little recent work done with comparison of biochemical properties of staphylococcal strains as a measure of pathogenicity, these strains were also screened for hemolysis production, mannitol fermentation, pigment production, and gelatinase production.

There was very little correlation observed among the biochemical properties, but hemolysis production and mannitol fermentation were most closely related to coagulase production. Furthermore, there was little correlation between the biochemical properties and bacteriophage typing.

Of the 11 antibiotics used in the sensitivity test, the strains proved to be most resistant to dihydrostreptomycin, phenoxyethyl penicillin and oxytetracycline. There was some correlation between resistance to antibiotics and phage typeability, with those typed at RTD correlating more closely than those typed at 100 X RTD.

Bacteriophages of the international series (human group), and some canine and bovine phages were used. Thirteen and one half per cent of the coagulase-positive strains were typed at

RTD while 20.1% were typed at 100 X RTD. Generally speaking, the canine phages were more specific for the canine isolates; however a greater number were typed by the international series.

The fact that some, if only a small per cent, of the isolates of canine origin were found to be typeable by the international series indicated that there are strains of Staphylococcus aureus common to both dog and man. The significance of the findings in this study is, of course, limited and further search will be necessary to determine the extent of interspecies relationship of staphylococcal infections.