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RESEARCH NOTE  
NON-GROWTH AT 45°C OF CERTAIN STRAINS OF  
CHICKEN *STAPHYLOCOCCUS AUREUS*

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*Staphylococcus aureus* from chickens, canaries, cows, pigs, horses, dogs and humans was tested for growth at 45°C. Chicken strains gave the lowest frequency of growth (33.2%). Strains of other sources grew with high frequency (95~100%). This low frequency in chicken *S. aureus* was due to the incidence of certain biological types, all strains of which could not grow at the incubation temperature.

INTRODUCTION

There have been descriptions of *Staphylococcus aureus* growing in or on appropriate nutrient media at 45°C<sup>1,2)</sup>. However, there has been no description on the response of chicken *S. aureus* to the incubation temperature.

Recently SATO et al. (1969) reported that the most of the chicken coagulase-positive staphylococci could be subdivided into 4 biological types on the basis of investigations into more than 1,000 strains from many flocks. The types are shown in table 1. Strains of type 2 were of unusual characteristics of *S. aureus*.

TABLE 1 *Biological types of chicken coagulase-positive staphylococci*\*<sup>1</sup>

BIOLOGICAL TYPE	PIGMENT	HEMOLYSIS ON SHEEP BLOOD AGAR PLATE	VOGES-PROSKAUER REACTION	LACTOSE AND GALACTOSE FERMENTATION	MANNITOL FERMENTATION
1	yellow	+	-	-* <sup>2</sup>	+
2	white	-	-	+	-
3	yellow	+	+	+	+
4	yellow	+	-	+	+

\*<sup>1</sup> SATO et al. (1969)

\*<sup>2</sup> Sometimes fermented after prolonged incubation

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This note deals with the non-growth at 45°C of certain biological types of the organisms of chicken origin.

#### MATERIALS AND METHODS

##### Sources of strains

**Chickens:** A total of 404 strains were employed in this study. The strains had been collected for the previous study<sup>3)</sup>. Three hundred and eighty-nine of them were isolated from diseased (dermatitis, septicemia, and arthritis) or from apparently healthy chickens (skin, oral cavity, nasal sinus, cloaca and eye) and from air samples in chicken houses. They were isolated in Hokkaido, and in the Tokyo and Osaka districts. The remaining 15 strains were derived from dead chicken embryos from 3 hatcheries in Hokkaido. The strains tested had been preserved in a cooked meat medium for a few years.

**Canaries:** Ten strains were isolated from the birds infected with pox. They were obtained in 2 localities in Hokkaido.

**Cows:** A total of 152 strains examined were isolated mainly from the udders of healthy milking cows in Sapporo and Ebetsu, Hokkaido.

**Pigs and horses:** Twenty-five and 50 strains respectively were isolated from the nasal cavities, tonsils or intestinal contents of healthy animals of both species, which were slaughtered at an abattoir in Sapporo.

**Dogs:** Sixty-one strains were derived from diseased (dermatitis, arthritis, metritis and canine distemper) and healthy dogs (skin, nasal cavity, eye and ear canal).

**Humans:** A total of 61 strains were employed. They were isolated from inpatients, doctors, nurses, and medical apparatus of 2 university hospitals (22 and 39 strains, respectively) in Sapporo.

##### Test for growth at 45°C

A loopful (loop diameter- 3.5 mm) of overnight broth culture of test strains was inoculated into 2 ml of trypticase soy broth (BBL) in a small test tube. Each tube was incubated at 45°C for 24 hr in a water bath. The strains were recorded as positive for the growth at 45°C, when the turbidity of media inoculated with them apparently increased as compared with a controlled uninoculated medium.

#### RESULTS AND DISCUSSION

As can be seen in table 2, chicken staphylococci grew at 45°C with lower frequency compared with the organisms from other sources.

Table 3 shows that the lower frequency of the growth in chicken staphylococci is due to the incidence of biological type 1, although also type 4 and a few strains of type 3 indicated non-growth.

SATO et al. (1969) investigated additional characteristics of each biological type such as DNase activity, the egg-yolk factor, the production of caseinase or fibrinolysin, susceptibility to the phages of international typing set and antigenic types of coagulase<sup>4)</sup>. DNase activity was positive in all the types, but

TABLE 2 *Growth at 45°C of S. aureus from human and animal sources*

SOURCE	NO. OF STRAINS TESTED	GROWTH AT 45°C	
		-	+
Chickens	404	270	134 (33.2%)
Canaries	10	0	10 (100%)
Cows	152	0	152 (100%)
Pigs	25	0	25 (100%)
Horses	50	2	48 (96%)
Dogs	61	3	58 (95.1%)
Humans	61	2	59 (96.7%)

TABLE 3 *Relation of growth at 45°C of chicken S. aureus to the biological types*

BIOLOGICAL TYPE	NO. OF STRAINS TESTED	GROWTH AT 45°C		
		-	+	Per cent of positives
1	261 (8)* <sup>1</sup>	261 (8)	0	0
2	37 (2)	0	37 (2)	100
3	101 (20)	4 (4)	97 (18)	96.6
4	5 (1)	5 (1)	0	0
Total	404 (27)* <sup>2</sup>	270 (13)	134 (20)	33.2

\*<sup>1</sup> No. of farms from which strains tested were isolated

\*<sup>2</sup> No. of farms

Multiple types were found in some farms.

the egg-yolk factor only in types 3 and 4. Production of caseinase and fibrinolysin was variable in each type. Thirty-eight % of type 3 strains were typable with the phages, and other strains tested were unsusceptible to them. All strains of types 1 and 4 belonged to antigenic type V of coagulase. The coagulase type did not occur in the other biological types. Furthermore, all strains of the biological types 1 and 4 could not grow at 45°C incubation as described above. These findings indicate that there seems to be a close relationship between the biological types found by SATO et al. and the growth ability at 45°C, as well as the egg-yolk factor or antigenic type of coagulase.

Thus the growth ability at 45°C of chicken *S. aureus* appears to be a valuable marker in the characterization of the organisms.

Since the body temperature of the birds is higher than that of the other

animals, it is interesting to know why certain strains of chicken *S. aureus* did not grow at 45°C. A study is needed to clarify this question.

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