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EXPERIMENTAL SALMONELLOSIS IN JAPANESE QUAILS

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Salmonellosis is an important disease of chicken and in many farms it appears in an endemic form. However, reports on salmonellosis in quails are scanty, from India. Experimental infection of turkeys, quails, and chicken has been reported by Adler *et al.* (1953), Edgar *et al.* (1964), Olesvik *et al.* (1969) and Brown *et al.* (1975). From India, salmonellosis in quail caused by *S. bareilly* and *S. gallinarum* was reported by Kapoor *et al.* (1980) and Sarma *et al.* (1988). *S. typhimurium* infection in Japanese quails was reported by Terada *et al.* (1988).

During the course of investigation on hepatic disorders in quails *S. typhimurium* was isolated from the liver of quails. In order to study the pathogenesis and pathology of salmonellosis in quails an experimental study was designed and the results obtained have been documented in this paper. Till now there has not been any report of *S. typhimurium* infection in quails from Kerala.

Materials and Methods

From the fresh carcasses of quails brought for autopsy to the Centre of Excellence in Pathology representative samples of liver with gross lesions were collected for bacteriological examination. Bacteriological cultures were made in Tryptose Soya Agar, MacConkey agar, and blood agar and incubated at 37°C for 24 h. The genera of the isolates were identified as described by Cowan, (1974). For systematic typing and identification the cultures were sent to the National Salmonella and Escherichia Centre, C.R.I., Kasuali.

From the fresh carcasses representative samples of liver, brain, kidney, spleen and bursa were collected in 10 per cent neutral buffered formalin for histopathological examination. The tissues were processed by routine methods, embedded in paraffin and sections cut at 3-5 u thickness were stained with hematoxylin and eosin.

Experimental design

Thirty, three-month-old healthy Japanese quails were randomly divided into five groups of six quails each and maintained in cages during the experimental investigation. Before the commencement of the experiment, all the birds were tested and found negative for *S. pullorum* and *S. gallinarum* infection by rapid plate test using pullorum coloured antigen obtained from the Institute of Animal Health and Veterinary Biologicals, Bangalore, India.

The inoculum used was a saline suspension of 18 h old Tryptose soya agar culture containing 10^8 organisms per ml, determined by McFarland Nephelometer

standards (Sonnenwirth, 1990). The bacteria were injected into the crop of the quails using a sterile tuberculin syringe. Group A was administered 0.3 ml of the suspension per bird; Group B was administered 0.6 ml of the suspension per bird; Group C was administered 0.9 ml of the suspension per bird; Group D was given cortisone acetate @ 6 mg/bird, IM, for three days to induce immunosuppression and on the fourth day 0.3 ml of the bacterial suspension per bird was administered, and Group E was kept as control. The control group was housed away from the treatment groups.

The quails were observed for fifteen days. The birds which died during the course of the experiment and the remaining birds which were sacrificed on the 15th day were subjected to detailed post-mortem examination and gross lesions were recorded. Representative samples of liver were collected for bacteriological examination and samples of liver, spleen, brain, kidney and bursa were collected in 10 per cent neutral buffered formalin for histopathological examination.

Results

Salmonella typhimurium was isolated from the liver of three quails during the course of the post-mortem examination of the quails brought for autopsy to the Centre of Excellence in Pathology.

Within 24 h of the administration of the bacterial suspension which was isolated from quails which died spontaneously, all the quails became slightly dull and showed more fluid consistency of the faecal droppings. All the infected quails became dull and suffered progressive loss in weight till the 8th day as against the steady weight gain noticed in the control birds (Table 1). Compared to the birds which did not receive corticosteroid but only bacterial suspension the corticosteroid treated quails became severely depressed within 24 h of the infection. Out of six corticosteroid treated birds, two birds died within 24 h and the remaining four birds died on the third day of the experimental infection with *S. typhimurium*. The birds from Group C died on the second day. One bird from the Group B developed torticollis and it died on the 5th day of the experiment. Remaining birds from all the groups were sacrificed on the 15th day.

Gross changes

The birds which died within 24 h of the treatment had moderate to severe congestion of the liver, kidney, spleen and heart. Two to three pin head sized greyish-white necrotic foci were seen on the liver. The birds which were sacrificed on 15th day had moderate hepatic congestion and few to many greyish-white foci of necrosis scattered throughout the liver. Spleen and heart were moderately congested. All the birds in group D died within 72 h of infection and liver from these birds were severely congested. Birds which died on second day had many scattered greyish-white foci and congestion of the spleen and heart. Liver of one bird from this group had numerous such small foci appearing like a saw dust liver. There was catarrhal enteritis in all birds, but it was more severe in quails which died naturally.

Table 1. Average body weight (mean ± SE) of quails in experimental salmonellosis (g)

Group	Days							
	1	2	3	4	5	6	7	8
A	91±0.73^{a}	90±0.58^{a}	88±1.10^{a}	89±1.10^{a}	90±1.50^{ab}	91±1.15^{a}	92±1.15^{b}	95±0.86^{a}
B	89±0.86^{a}	88±0.58^{a}	88±0.93^{a}	85±0.82^{b}	87±1.71^{ac}	90±0.52^{ab}	90±1.26^{a}	91±0.71^{ab}
C	89±0.86^{a}	84±1.15^{b}	83±1.78^{b}	84±0.86^{c}	87±1.21^{bc}	88±0.63^{b}	90±0.71^{a}	91±1.10^{b}
Control	90±0.97^{a}	98±0.93^{c}	98±0.93^{c}	104±1.71^{c}	112±1.39^{d}	115±1.03^{d}	118±0.73^{d}	121±1.53^{d}

Means with common letters do not differ significantly

Histologic changes

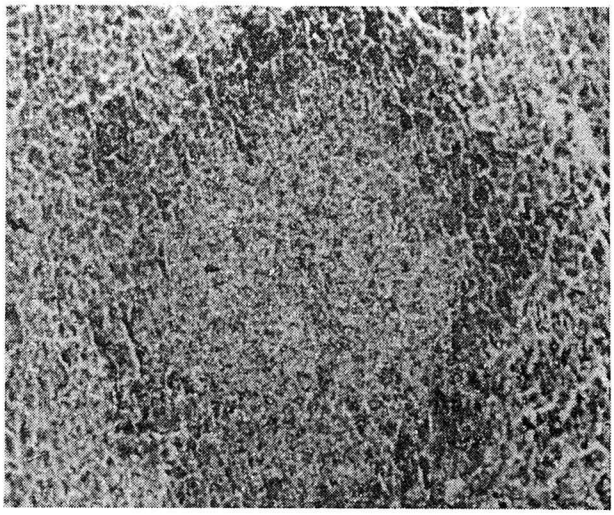

Fig. 1 Nocrotic focus in the liver parenchyma. Compressed hepatocytes corrounding the zone of heterophils and necrosis. H&E x 400

The histologic changes were typical of bacterial hepatitis. There were focal to multifocal areas of coagulative necrosis of the liver. In focal areas infiltration by heterophils was evident. The liver from group C had extensive infiltration of heterophils. Compressed hepatocytes were seen surrounding this zone (Fig. 1). In severely congested cases hepatic sinusoids were stuffed with erythrocytes, otherwise there was localized dilatation of capillaries. In 18 of the 24 cases there was moderate to severe fatty change in the liver. All the corticosteroid treated birds had severe

congestion, multifocal areas of necrosis and moderate fatty change. Two of the six birds in this group had infiltration of heterophils in focal areas in addition to the above changes. Here the infiltration was mostly perivascular.

In the spleen there was reticular hyperplasia and moderate to severe congestion. In the heart there was mild myocardial degeneration, and capillaries were engorged. Brain and kidney had mild to moderate congestion. All the infected quails had catarrhal enteritis. Liver and other organs from the control birds did not reveal pathological alterations.

Discussion

This investigation has established that salmonellosis is prevalent in quails and it was also clarified that the organism involved was *S. typhimurium*. Experimentally it was found that quails could be infected with strains of *S. typhimurium* isolated from spontaneous cases (Edgar *et al.*, 1964). It was found in this study that stress played a significant part in enhancing the pathogenicity of the organism. In experimentally induced stress, the quails were found to be very susceptible, the distribution of lesions was extensive and mortality was high and quicker. This implies that in natural infections, when quails are under stress they will pick up the infection easily. Therefore, there is need to prevent stress during rearing of quails. The observations made in this study were similar to those observed by Brown *et al.* (1975) in oral *S. typhimurium* infection in chicken. Terada *et al.* (1988) isolated *S. typhimurium* from Japanese quails but the details of the experiment were not available.

There was progressive loss of body weight till the 8th day, after that there was improvement but the quails failed to gain optimum body weight till the 15th day. Early infection was followed by a transient bacteraemia. These observations support the findings of Brown *et al.* (1975).

Brown *et al.* (1975) observed the most significant lesions in the intestinal tract with inflammation of the duodenum, upper ileum and caeca in cockerels infected with *S. typhimurium*. But in the present case, quails which died naturally had severe catarrhal enteritis but the quails which were sacrificed on the 15th day had only mild catarrhal enteritis.

The gross distribution of lesions and the perivascular location of the lesions indicated a bacteraemic phase and localization of the organisms in the liver. The necrotic foci were the classical lesion found in salmonellosis and was similar to those observed in chicken. The immunosuppression, naturally caused more extensive and severe lesions.

S. typhimurium was isolated from the liver upto 15th day. This was in contrast to the observations made by Brown *et al.* (1975) in chicken. They were able to isolate the organisms from cockerels infected with *S. typhimurium* upto 5 days, and after that a fall in the recovery rate occurred.

Summary

Salmonella typhimurium was isolated from three adult quails which had died spontaneously. Experimental infection was produced in healthy quails and after immunosuppression with cortisone acetate. The lesions were severe and extensive in immunosuppressed quails. Induced stress was found to enhance the pathogenicity of the organism. The role of stress in precipitating the disease in field situation was thus clarified. Lesions were seen predominantly in the liver and intestines. Hepatic necrosis was a consistent feature.

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