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ASCORBIC ACID, CAROTENOIDS AND VITAMIN
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WITH CORYZA, CHOLERA OR
NEWCASTLE DISEASE

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R. L. SQUIBB, J. E. BRAHAM, M. GUZMAN AND N. S. SCRIMSHAW

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Blood Serum Total Proteins, Riboflavin, Ascorbic Acid, Carotenoids and Vitamin A of New Hampshire Chickens Infected with Coryza, Cholera or Newcastle Disease

ROBERT L. SQUIBB,¹ J. EDGAR BRAHAM,² MIGUEL GUZMAN³ AND NEVIN S. SCRIMSHAW,³

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EACH year in Central America epidemics of coryza, cholera or Newcastle disease singly or in combination significantly reduce the poultry population. Available drugs or vaccines have not proven effective in controlling coryza or cholera infections. Therefore, additional basic knowledge and control measures are needed for the treatment and especially for the prevention of these diseases. Newcastle disease, on the other hand, may be controlled with the use of vaccines.

Outbreaks of coryza and cholera during October 1953 to May 1954, in the New Hampshire flocks of the Instituto Agropecuario Nacional (IAN) and Newcastle disease in a local New Hampshire flock

made it possible to study the effect of these diseases on blood serum levels of total proteins, riboflavin, ascorbic acid, total carotenoids and vitamin A of infected birds.⁴ Limited serum amylase determinations also were made on birds infected with cholera.

EFFECT OF CORYZA ON BLOOD SERUM CONSTITUENTS

Materials and Methods.—The disease coryza was studied over a 6 month period during which blood samples were obtained from 5 flocks of New Hampshire chickens of different ages and sex. At the time the blood samples were obtained, four of the flocks had a moderate infection of coryza, characterized by a swelling of the head accompanied by a thick, sticky nasal discharge. The birds of a fifth flock were considered to have an advanced infection since they not only showed these symptoms but also were unable to walk. All birds in this group died within 72 hours after blood samples were obtained.

Each time infected birds of one of the 5 flocks were bled, a similar set of samples was obtained from comparable healthy birds. These controls were of the same age and sex and were housed with the infected birds. Even though the controls did not show any clinical signs of coryza, each bird was observed for an additional one-week period. If any had contracted the disease, the resulting blood values would have been eliminated from the final comparisons.

All blood samples were drawn from the

¹ Instituto Agropecuario Nacional (IAN), a technical agricultural organization operated jointly by the Government of Guatemala and the Foreign Operations Administration of the United States of America.

² Research fellow of the Instituto de Nutrición de Centro América y Panamá and the Instituto Agropecuario Nacional.

³ Instituto de Nutrición de Centro América y Panamá (INCAP), human nutrition institute supported by the governments of Central America and Panama and administered by the Pan American Sanitary Bureau, Regional Office of the World Health Organization.

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Part of the data on the effect of coryza on blood serum levels were presented at the Federation Meetings in Atlantic City, April 12 to 17, 1954 (Fed. Proc. 13, 1954).

TABLE 1.—*Effect of coryza on blood serum total proteins, riboflavin, ascorbic acid, carotenoids and vitamin A of New Hampshire chicken*

Flock No.	Number of birds	Sex	Age	Stage of disease	Total prot.	Ribo-flavin	Ascorbic acid	Vit. A	Total carotenoids
			months		gm. %	mcg. %	mg. %	mcg. %	mcg. %
1	15	F	3	control	4.06	1.34	1.60	42	265
	15	F	3	moderate	4.59*	1.11	1.31	24†	158*
2	15	M	3	control	4.22	1.75	1.79	37	275
	15	M	3	moderate	4.82*	1.13*	1.18	21†	209
3	12	F	5	control	4.47	1.95	2.09	50	425
	12	F	5	moderate	4.38	1.53*	1.48†	31†	364
4	10	M	10	control	4.67	1.24	1.81	25	277
	10	M	10	moderate	4.86	1.66	1.75	17	240
5	10	F	10	control	4.58	0.82	2.08	35	279
	10	F	10	advanced	4.67	1.53†	1.53*	8†	241
Total Controls	62	—	—	—	4.36	1.42	1.87	39	303
Infected	62	—	—	—	4.66*	1.39	1.45†	21†	237*

* Significant at 5% level.

† Significant at 1% level.

large wing veins, allowed to clot and centrifuged immediately afterwards. The sera thus obtained were analyzed for: total proteins (Lowry and Hunter, 1945), ascorbic acid (Lowry *et al.*, 1945), riboflavin (Burch *et al.*, 1948), and total carotenoids and vitamin A (Bessey *et al.*, 1946).

Results.—The number of birds observed in each flock, their sex and age are presented in Table 1 together with the data from analyses of the sera.

In all five flocks, total carotenoids and ascorbic acid were depressed in the infected birds, however, this depression was significant only for the total carotenoids of flocks 1 and the serum ascorbic acid of flocks 3 and 5. Vitamin A was reduced significantly in flocks 1, 2, 3 and 5 and depressed in flock 4. When the data of the five flocks are pooled and treated statistically, regardless of sex, age or stage of infection, a significant reduction of serum ascorbic acid, total carotenoids and vitamin A occurred in coryza.

In four of the five flocks the serum protein levels are higher in infected birds than in those without coryza although the difference is significant only in flocks 1 and 2. In flock 3 serum protein values are slightly but not significantly lower in infected birds. The relative values for riboflavin in control and infected birds varied from flock to flock. When the data for all five flocks were pooled, no change in riboflavin values was apparent, but the elevation of serum protein in chickens with coryza was found to be significant.

EFFECT OF CHOLERA ON BLOOD SERUM CONSTITUENTS

Materials and Methods.—The effect of cholera on the several blood serum constituents was observed in three different flocks of New Hampshire chickens of different ages and sex. The spread and severity of the disease was so great that it was impossible to obtain blood samples from apparently normal birds. Therefore, in the cholera infected flocks the birds

TABLE 2.—*Effect of cholera on blood serum total proteins, riboflavin, ascorbic acid, carotenoids and vitamin A of New Hampshire chicken*

Flock No.	Number of birds	Sex	Age	Stage of disease	Total prot.	Ribo-flavin	Ascorbic acid	Vit. A	Total carotenoids
					gm. %	mcg. %	mg. %	mcg. %	mcg. %
6	9	¹	3 weeks	moderate	3.24	4.11	3.61	42	366
	9	¹	3 weeks	advanced	2.50†	4.73	3.97	26†	157†
7	8	M	6 mon.	moderate	4.52	1.93	2.24	35	171
	7	M	6 mon.	advanced	5.94*	1.05*	1.75	26	169
8	8	F	6 mon.	moderate	4.30	2.02	2.52	26	—
	11	F	6 mon.	advanced	7.00†	1.72	1.87	24	251
Total Controls ²	30	—	6 mon.	—	4.14	1.55	1.70	39	270
Infected	34	—	6 mon.	—	5.56†	1.70	2.04*	28†	177*

¹ Sex not determined.² Taken from flocks 2, 3, 4 and 5 of the coryza infected birds, Table 1.

* Significant at 5% level.

† Significant at 1% level.

were separated arbitrarily into the following 2 groups: 1) moderate infection, characterized by described clinical symptoms of the disease in birds still able to move around, 2) advanced infection, identified by the same clinical symptoms, in birds unable to walk. Advanced infection usually resulted in mortality within 48 hours.

Blood samples from infected birds were obtained by heart puncture, allowed to clot and were centrifuged immediately afterwards. All sera were analyzed for the blood constituents previously listed. In addition, a small number of serum samples from both normal and cholera infected birds of two different flocks were examined for their amylase activity by the method of King (1951).

Results.—The age and sex of the birds and the results of the analyses of the blood sera are presented in Table 2. These data show that in the 3 week old chicks (flock 6), total serum proteins, vitamin A and total carotenoids were significantly reduced as the disease became more advanced. Serum riboflavin and ascorbic acid, on the other hand, were slightly increased. In 6 month old birds

(flocks 7 and 8), total serum proteins were increased significantly as the disease became advanced, while riboflavin, ascorbic acid, carotenoids and vitamin A were reduced.

When blood serum constituent levels of control birds (flocks 2 and 3) were compared with infected birds (both moderate and advanced of flocks 7 and 8) it was found that serum total protein and ascorbic acid were significantly elevated while vitamin A and total carotenoids were reduced significantly. Table 3 shows that serum amylase values were reduced as the result of cholera infection in both 3 week old chicks and adult birds.

EFFECT OF NEWCASTLE DISEASE ON BLOOD SERUM CONSTITUENTS

Materials and Methods.—Severe outbreaks of Newcastle disease in 2 out of 3 flocks of 6 week old New Hampshire chicks on a nearby poultry farm made it possible to obtain a series of blood samples from: 1) a group of apparently non-infected birds, 2) a group suffering rapid mortality and 3) a group which were survivors of a previous attack. The birds

TABLE 3.—*Effect of cholera on the serum amylase activity of New Hampshire chicken*

Flock	Number of birds	Sex	Age	Stage of disease	Serum amylase	
					Average	Range
9	16	M. F.	3 wks.	control	units ¹ 2,168	units 1,231–3,556
	9	M. F.	3 wks.	advanced	1,520	508–1,391
10	8	F	12 mon.	control	1,243	744–1,600
	3	F	12 mon.	advanced	428	311– 508

¹ Amount of amylase which hydrolyzes 1.5 mg. of starch in 8 minutes.

of the latter group showed typical symptoms of "crazy chick" disease.

Approximately 3 ml. of blood were obtained by heart puncture from each of the birds. The blood was allowed to coagulate and the sera analyzed for the previously described blood constituents.

Results.—The number of birds sampled, the stage of disease and the resulting levels of the blood serum constituents are presented in Table 4. Total protein, vitamin A and total carotenoids were significantly reduced and ascorbic acid significantly elevated as result of the infection. No effect was observed on serum riboflavin. In the birds with crazy chick symptoms total serum protein was significantly elevated while serum ascorbic acid, vitamin A and total carotenoids were not significantly different from the control birds.

DISCUSSION

The results of the studies reported here leave no doubt that significant changes in the blood serum levels of vitamin A, carotenoids, ascorbic acid and total protein do occur in poultry infected with coryza, cholera or Newcastle disease.

Coryza, as observed in the New Hampshire flocks of the IAN, often lasts 4 to 8 weeks before death of the bird ensues. During this extended period, appetite and consequently feed intake are drastically reduced. While a significant elevation of total serum proteins and a depression of serum ascorbic acid, carotenoids and vitamin A was observed, levels of these blood constituents may have been influenced in part by the reduced feed intakes. Since serum carotenoid levels were relatively much less depressed in birds with coryza than levels of vitamin A, it is

TABLE 4.—*Total serum protein, riboflavin, ascorbic acid, vitamin A and carotenoids of New Hampshire chicks infected with Newcastle disease*

Flock	Number of birds	Age	Stage of disease	In 100 ml. serum				
				Total protein	Riboflavin	Ascorbic acid	Vitamin A	Total carotenoids
		<i>weeks</i>		<i>gm.</i>	<i>mcg.</i>	<i>mg.</i>	<i>mcg.</i>	<i>mcg.</i>
11	15	6	control	4.03	3.20	1.23	28.8	361
12	15	6	highly infected	3.53	3.33	1.89	18.1	148
13	16	6	"crazy chick"	4.55	3.58	1.57	34.4	287

Control and highly infected groups: Difference between total proteins, vitamin A and ascorbic acid significant at 5% level; total carotenoids, 1% level.

Highly infected and crazy chick stage: Difference between vitamin A and total carotenoids significant at 5% level, total protein, 1% level.

Control and crazy chick stage: Difference between total protein significant at 5% level.

possible that there was an interference with carotenoid conversion to vitamin A.

In the case of cholera infection, which causes rapid mortality, little or no effect of reduced feed intake on blood constituents can be postulated. Nevertheless, significant effects of the disease on serum amylase activity were observed as well as on total serum protein, vitamin A and carotenoid levels. The effect of cholera on serum ascorbic acid and riboflavin was highly variable.

It is recognized that experimental conditions were not the same between the flocks studied, however, certain effects on blood serum constituents were common to the 3 diseases. Serum riboflavin was not affected while serum total carotenoids and vitamin A were depressed by coryza, cholera and Newcastle disease. The other constituents varied with the specific disease. Total serum protein was elevated in coryza and cholera and reduced in birds infected with Newcastle. Serum ascorbic acid, on the other hand, was elevated in birds infected with cholera and depressed in birds infected with coryza.

Whether the restricted feed intake of birds with Newcastle infection had a direct influence on the blood constituents studied is not known.

The effect of coryza and cholera, two diseases for which there is no adequate control, on several serum blood constituent levels of infected birds suggest that the value of nutritional measures in the prophylaxis and therapy merit further study.

SUMMARY

A coryza infection in New Hampshire chickens of different ages and sex resulted

in a significant reduction of serum ascorbic acid, total carotenoids and vitamin A. Serum protein levels were found to be elevated in infected birds while little or no effect was observed on serum riboflavin levels.

Cholera infections were observed to result in rapid mortality. Serum total protein and ascorbic acid levels were found to be elevated while total carotenoids and vitamin A serum levels were significantly depressed. A depression of serum amylase activity in infected birds was also observed.

Newcastle disease significantly reduced total serum protein, vitamin A and carotenoids and elevated serum ascorbic acid. No effect was observed on serum riboflavin levels.

The findings suggest that the value of nutritional measures in the therapy and prophylaxis of birds infected with coryza and cholera should be investigated.

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