Effect of Amino Acid Injection in Broiler Breeder Eggs on Embryonic Growth and Hatchability of Chicks

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ABSTRACT Two experiments were conducted to evaluate the effect of amino acid (AA) injections *in ovo* in Cobb broiler breeder eggs on hatchability and subsequent chick BW. In Experiment 1, moisture, crude fat (CF), and CP were analyzed over time during incubation (Day 0, 7, 14, and 19 of incubation). Moisture, CP, and CF of the embryo increased, and moisture, CP, and CF of eggs decreased, as incubation time increased ($P < 0.05$). Combined egg and embryo AA contents, except Gly and Pro, decreased ($P < 0.05$) as incubation time increased. However, the pattern of AA in the egg did not change as the embryo developed. In Experiment 2, AA were injected into the yolk or air cell at Day 0 and 7 of incubation. Hatchability was reduced ($P < 0.05$) when AA were injected at Day 0 of incubation. However, when the AA solution was injected into the yolk sac at Day 7 of incubation, hatchability was not affected, and BW of chicks increased relative to egg weight prior to incubation. These results suggest that *in ovo* administration of AA may be an effective method of increasing chick BW at hatch.

(*Key words*: amino acid, *in ovo*, egg, hatchability, broiler breeder)

1999 Poultry Science 78:1493–1498

INTRODUCTION

Large eggs have larger embryos than embryos from smaller eggs (Al-Murrani, 1978). A 1-g difference in egg weight results in a 10-g difference in 56-d broiler BW (Al-Murrani, 1978). Thus, broiler breeder eggs at the beginning of egg production may result in chicks of low BW. In contrast to mammals, avian embryo development is restricted by the nutrients within the egg. Also, embryo and chick weight are influenced by yolk nutrient status because of the chicks' yolk utilization posthatch.

Broiler breeder eggs contain an excess of fat and moisture but not protein (Al-Murrani, 1978). It has been shown, however, that broiler breeders fed low energy-high protein diets have eggs with decreased hatchability (Pearson and Herron, 1982). Al-Murrani (1982) demonstrated that injecting an amino acid (AA) mixture (identical to the AA pattern of egg protein) into growing embryos of broiler breeder eggs resulted in higher chick BW at hatch and at 56 d of age as compared with chicks from control embryos. The US poultry industry's acceptance of *in ovo*-administered vaccination programs is increasing (Johnston *et al.*, 1997). However, research concerning *in ovo* nutrient administration (*e.g.*, AA) in broiler breeder eggs is minimal. The present study was conducted to evaluate *in ovo* administration of AA on embryos and subsequent chick weight in addition to monitoring embryonic changes in CP, AA, crude fat (CF), and moisture content during incubation.

MATERIALS AND METHODS

Two experiments were conducted utilizing 250 fertilized commercial eggs of a Cobb strain. All eggs were obtained from the same breeder flock and were laid within a 24-h period. Eggs were incubated at 37.8 C and 60% RH.

Experiment 1 was conducted to evaluate the transfer of nutrients from the egg to the embryo at different days of incubation. One hundred eggs were selected randomly and divided into four groups of 25 eggs, each weighing an average of 73.0 ± 0.3 g. On Days 0, 7, 14, and 19 of incubation, 20 eggs from each group were weighed, and contents of egg and embryo were collected. Embryos were removed from the amniotic fluid and separated from the yolk sac. Five embryos were pooled, to give the required mass for analysis, and minced with a mincing machine. Samples were freeze-dried until analyzed for CP, CF, moisture, and AA. The CP content of egg and embryos was determined by the micro-Kjeldahl method, CF was determined by ether extraction, and moisture was determined by drying at 105 C for 24 h according to AOAC

Received for publication June 29, 1998.
Accepted for publication July 1, 1999.
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Abbreviation key: AA = amino acid, CF = crude fat.

TABLE 1. Composition of amino acid (AA) solution injected into hatching eggs

AA	Content[1]	Pattern
	(mg/0.5 mL)	(% Lys)
Aspartic acid	5.31	140.5
Threonine	2.53	66.9
Serine	3.86	102.1
Glutamic acid	6.99	184.9
Glycine	1.77	46.8
Alanine	3.01	79.6
Valine	3.34	88.4
Cysteine	1.10	29.1
Methionine	1.91	50.5
Isoleucine	2.71	71.7
Leucine	4.53	119.8
Tyrosine	1.84	48.7
Phenylalanine	2.81	74.3
Lysine	3.78	100.0
Histidine	1.35	35.7
Arginine	3.24	85.7
Proline	1.96	51.9
Tryptophan	0.95	25.1
Total	53.0	

[1]Values calculated from results in Experiment 1.

(1980). All AA except Met and Cys were determined with an AA analyzer (HITACHI L-8500[2]) after hydrolysis with 6N HCl at 110 C for 22 h. Both Met and Cys were oxidized with performic acid before hydrolysis (Slumys and Bos, 1985).

In Experiment 2, a 0.5-mL AA solution that matched the AA pattern of an egg was injected into the yolk sac or air cell in 15 eggs weighing an average of 68.7 ± 3.8 g on Days 0 and 7 of incubation. Al-Murrani (1978) suggested that differences in protein content of eggs at Day 7 of incubation affect the growth of embryos. In addition, Vitamin D precursor administration into eggs has been done from Day 0 of incubation in Japanese quail (Elaroussi *et al.*, 1993). If AA administration is possible before incubation, it may be safer. Therefore, AA were administered on Day 0 and 7 of incubation. The large end of the egg (injection site) was sterilized prior to incubation with 70% ethanol. Egg injection treatments consisted of 0.5 mL of AA solution, 0.5 mL of sterile water, or no injection (control). A 1-mL disposable syringe and a 22-gauge needle were used for treatment injections into the eggs. The 0.5-mL injection solution (Table 1) contained the same amount of AA as that utilized by the growing embryo during 7 d as described by Al-Murrani (1982). The Trp concentration was calculated from a Trp:Lys ratio in the egg (Long, 1971). An injection solution of 0.1 mL solution was injected five times at 24-h intervals because the air cell was too small to be injected with 0.5 mL solution at one time on Day 0 of incubation. Needle punctures in the shell were sealed immediately with paraffin wax. After injection, eggs were returned to the incubator, and the BW of hatched chicks was recorded.

Treatments were analyzed by ANOVA using the General Linear Models procedure of SAS® (SAS Institute, 1988). When differences among means were found, means were separated using the LSD test. Statements of significance are based on $P < 0.05$ unless otherwise indicated. The comparison of hatchability data was done by the chi-square test.

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TABLE 2. Embryo, egg, and shell weights of broilers during incubation[1]

Days of incubation	Embryo	Egg	Shell	Total
	(g)			
0	. . .	66.4 ± 2.5[a]	7.0 ± 0.6[ab]	73.4 ± 2.7[a]
7	1.0 ± 0.2[a]	61.7 ± 2.3[b]	7.2 ± 0.5[a]	69.9 ± 2.6[b]
14	15.4 ± 0.9[b]	44.7 ± 2.0[c]	6.8 ± 0.5[bc]	66.9 ± 2.1[c]
19	38.4 ± 2.2[c]	19.9 ± 2.5[d]	6.6 ± 0.5[c]	64.9 ± 2.4[d]

[a–d]Means in the same column with no common superscripts differ ($P < 0.05$).

[1]Values are means ± SD for 20 embryos, eggs, and shells.

RESULTS

Experiment 1

Throughout incubation, embryo weight increased, and shell weight decreased (Table 2). Egg shell weight tended to decrease after Day 7 of incubation. The sum of embryo, egg, and shell weights decreased to 88% of initial weight at Day 19 of incubation.

In comparison with CP, moisture content was higher, and CF was lower, in eggs and embryos at all stages of incubation (Table 3). Egg CP, CF, and moisture decreased, and embryo CP, CF, and moisture increased, over time during incubation. The change in CP, CF, and moisture relative to the initial egg is shown in Figure 1. Total contents of CP did not change, moisture decreased to 90%,

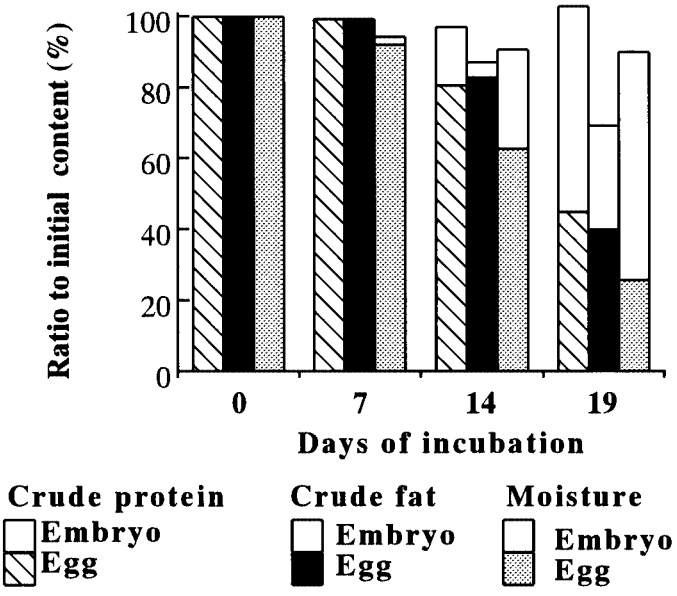

FIGURE 1. Time course of ratios of crude protein, crude fat, and moisture in egg and embryo of broilers to those before incubation. Values are means of 20 eggs.

TABLE 3. Protein, fat, and moisture contents of egg and embryo of broilers[1]

Days of incubation	Crude protein		Crude fat		Moisture	
	Egg	Embryo	Egg	Embryo	Egg	Embryo
	(g)					
0	8.88 ± 1.49[a] (52.23)[2]	...	5.81 ± 0.92[a] (34.17)	...	49.4 ± 3.6[a]	...
7	8.78 ± 0.58[a] (54.53)	0.06 ± 0.01[c] (85.71)	5.76 ± 0.41[a] (35.78)	0.01 ± 0.01[c] (14.29)	45.6 ± 1.9[a]	0.9 ± 0.0[c]
14	7.20 ± 0.55[b] (53.73)	1.43 ± 0.11[b] (84.12)	4.83 ± 0.45[b] (36.04)	0.24 ± 0.04[b] (14.12)	31.3 ± 1.7[c]	13.7 ± 0.9[b]
19	4.04 ± 0.50[c] (57.71)	5.15 ± 0.35[a] (74.64)	2.33 ± 0.55[c] (32.86)	1.71 ± 0.11[a] (24.78)	12.9 ± 2.0[d]	31.5 ± 1.8[a]

[a–c]Means in the same column with no common superscript differ ($P < 0.05$).
[1]Values are means ± SD for 20 eggs and five replications of four embryos.
[2]Percentage of dry matter.

and total CF decreased exponentially to 69% as incubation time increased. The ratios of transferred CP, CF, and moisture into the embryo were 58, 29, and 64% of egg, respectively, on Day 19 of incubation. Also, the transfer ratio of CF was one-half the level of other nutrients.

All AA of egg decreased as incubation time increased. The total AA contents of egg and embryo decreased, except Gly and Pro, as incubation time increased (Table 4). The AA contents of egg and embryo as a function of Lys are shown in Figure 2. Because the ideal protein of broilers is described as the AA ratio to Lys, the aminograms in egg and embryo are described as ratios to Lys (Baker and Han, 1994). Essential AA pattern of egg was constant,

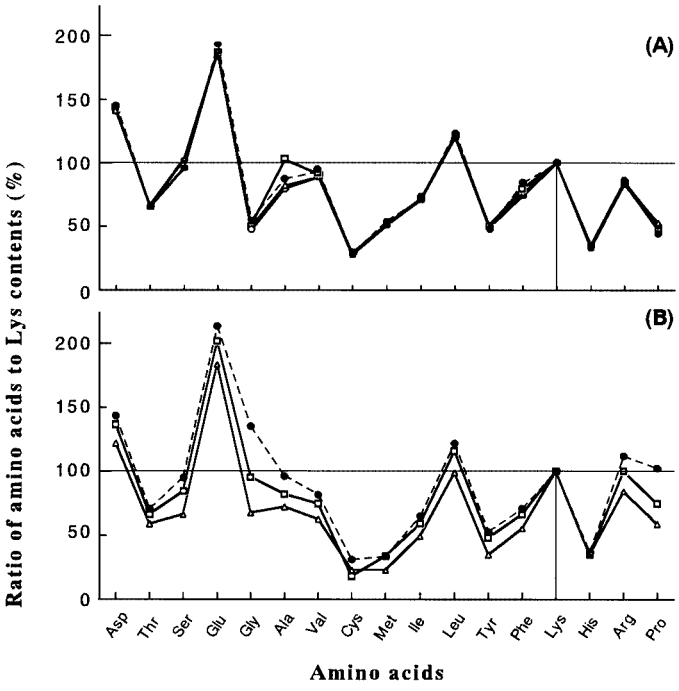

FIGURE 2. Aminogram of egg content (A) and embryo (B) on Days 0 (○), 7 (□), 14 (△), and 19 (●) of incubation. Each value is a ratio to lysine (Lys) contents (Table 4). Asp = aspartic acid; Thr = threonine; Ser = serine; Glu = glutamic acid; Gly = glycine; Ala = alanine; Val = valine; Cys = cystine; Met = methionine; Ile = isoleucine; Leu = leucine; Tyr = tyrosine; Phe = phenylalanine; Lys = lysine; His = histidine; Arg = arginine; and Pro = proline.

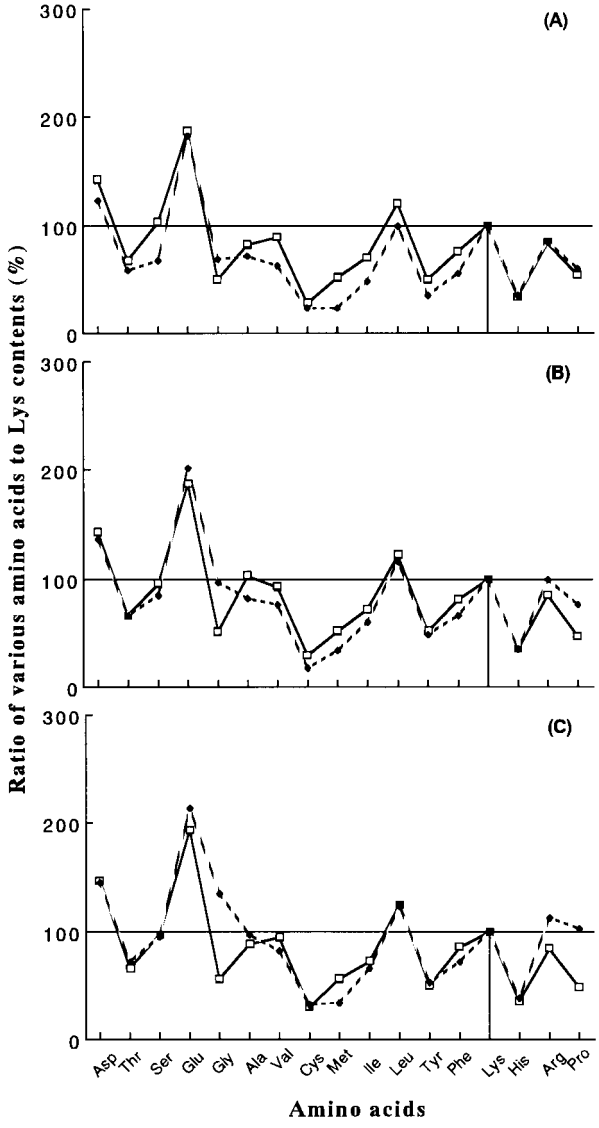

FIGURE 3. Comparison of aminogram in egg (□) and embryo (◆) on Day 7 (A), 14 (B), and 19 (C) of incubation. Each value is a ratio to lysine contents (Table 4). Asp = aspartic acid; Thr = threonine; Ser = serine; Glu = glutamic acid; Gly = glycine; Ala = alanine; Val = valine; Cys = cystine; Met = methionine; Ile = isoleucine; Leu = leucine; Tyr = tyrosine; Phe = phenylalanine; Lys = lysine; His = histidine; Arg = arginine; and Pro = proline.

but Gly and Pro increased in embryos with advancing incubation time. The aminograms described as ratios of each AA to Lys in egg and embryo on Day 7, 14, and 19 are shown in Figure 3. The AA ratios of embryo and egg were similar.

Experiment 2

Because the pattern of AA transferred to embryo was similar to that of the egg in Experiment 1, the AA solution with the same AA pattern as an egg was prepared for

TABLE 4. Time course of amino acid (AA) contents in the embryo and egg of broilers during incubation[1]

		Days of incubation			
	AA[2]	0	7	14	19
		(mg)			
Egg	Asp	907.9 ± 28.2[a]	811.1 ± 51.1[b]	735.6 ± 77.1[c]	404.3 ± 34.7[d]
	Thr	432.3 ± 14.3[a]	380.7 ± 25.9[b]	33.97 ± 36.0[c]	183.1 ± 12.9[d]
	Ser	659.4 ± 22.0[a]	588.8 ± 47.5[b]	495.1 ± 50.0[c]	266.6 ± 16.2[d]
	Glu	1,195.3 ± 33.8[a]	1,073.7 ± 72.4[b]	972.4 ± 96.5[c]	536.4 ± 37.5[d]
	Gly	302.9 ± 11.0[a]	280.7 ± 13.4[a]	265.0 ± 30.7[b]	153.9 ± 14.7[b]
	Ala	515.4 ± 17.2[a]	471.2 ± 25.7[a]	530.4 ± 177.2[a]	244.4 ± 17.7[b]
	Val	570.4 ± 14.8[a]	506.1 ± 26.1[b]	473.7 ± 47.6[b]	259.1 ± 22.3[c]
	Cys	187.9 ± 15.5[a]	158.8 ± 8.6[a]	151.4 ± 25.6[a]	80.0 ± 10.4[b]
	Met	325.9 ± 14.8[a]	290.0 ± 16.9[b]	267.6 ± 30.5[b]	152.1 ± 13.5[c]
	Ile	463.4 ± 10.2[a]	405.4 ± 23.8[b]	371.5 ± 38.7[c]	197.9 ± 18.3[d]
	Leu	773.9 ± 20.3[a]	686.3 ± 41.1[b]	628.4 ± 63.6[c]	345.1 ± 26.0[d]
	Tyr	315.4 ± 7.7[a]	286.0 ± 13.0[b]	262.9 ± 36.0[b]	136.8 ± 12.7[c]
	Phe	481.2 ± 14.1[a]	433.4 ± 30.8[b]	414.6 ± 41.6[b]	234.4 ± 18.0[c]
	Lys	645.7 ± 19.7[a]	571.3 ± 36.5[b]	517.6 ± 51.3[c]	277.0 ± 24.7[d]
	His	231.3 ± 9.6[a]	190.8 ± 19.0[b]	180.5 ± 22.4[b]	97.0 ± 10.4[c]
	Arg	553.9 ± 15.3[a]	480.6 ± 33.9[b]	439.5 ± 46.4[b]	230.5 ± 18.6[c]
	Pro	334.9 ± 17.2[a]	306.0 ± 20.0[a]	244.6 ± 31.4[b]	133.5 ± 13.2[c]
	Total	8,897.0 ± 269.1[a]	7,921.0 ± 457.4[b]	7,290.4 ± 772.1[b]	3,932.2 ± 309.0[c]
Embryo	Asp	...	4.0 ± 0.2[c]	102.4 ± 7.9[b]	356.2 ± 15.6[a]
	Thr	...	1.9 ± 0.1[c]	49.5 ± 4.5[b]	176.8 ± 9.6[a]
	Ser	...	2.2 ± 0.2[c]	63.5 ± 5.9[b]	235.8 ± 14.6[a]
	Glu	...	6.0 ± 0.4[c]	151.7 ± 11.4[b]	528.4 ± 26.4[a]
	Gly	...	2.2 ± 0.3[c]	71.9 ± 6.7[b]	333.1 ± 23.6[a]
	Ala	...	2.4 ± 0.2[c]	61.9 ± 5.0[b]	238.3 ± 8.0[a]
	Val	...	2.1 ± 0.1[c]	56.7 ± 4.5[b]	202.4 ± 8.9[a]
	Cys	...	0.8 ± 0.2[c]	13.2 ± 1.7[c]	77.7 ± 9.5[b]
	Met	...	0.7 ± 0.3[c]	25.7 ± 1.8[b]	82.8 ± 4.8[a]
	Ile	...	1.6 ± 0.1[c]	44.7 ± 3.3[b]	161.8 ± 5.8[a]
	Leu	...	3.2 ± 0.2[c]	87.1 ± 6.8[b]	302.2 ± 13.3[a]
	Tyr	...	1.2 ± 0.1[c]	36.4 ± 3.1[b]	130.2 ± 9.2[a]
	Phe	...	1.8 ± 0.2[c]	49.5 ± 4.1[b]	176.4 ± 6.5[a]
	Lys	...	3.3 ± 0.2[c]	75.0 ± 5.9[b]	246.7 ± 14.5[a]
	His	...	1.1 ± 0.1[c]	26.7 ± 2.3[b]	93.0 ± 3.8[a]
	Arg	...	2.8 ± 0.2[c]	75.0 ± 6.2[b]	278.1 ± 16.9[a]
	Pro	...	1.9 ± 0.3[c]	56.6 ± 5.5[b]	252.9 ± 19.3[a]
	Total	...	39.1 ± 2.9[c]	1,047.4 ± 83.9[b]	3,872.8 ± 188.5[a]
Total	Asp	907.9 ± 28.2[a]	815.1 ± 51.3[bc]	837.9 ± 77.5[ab]	760.5 ± 47.6[c]
	Thr	432.3 ± 14.3[a]	382.6 ± 26.0[b]	389.2 ± 35.7[b]	360.0 ± 20.7[b]
	Ser	659.4 ± 22.0[a]	591.0 ± 47.6[b]	558.7 ± 48.3[b]	502.4 ± 27.5[c]
	Glu	1,195.3 ± 33.8[a]	1,079.7 ± 72.7[b]	1,124.1 ± 95.7[ab]	1,064.7 ± 57.0[b]
	Gly	302.9 ± 11.0[c]	282.9 ± 13.6[c]	336.9 ± 29.3[b]	487.0 ± 31.1[a]
	Ala	515.4 ± 17.2	473.5 ± 25.9	592.3 ± 178.8	482.6 ± 25.0
	Val	570.4 ± 14.8[a]	508.1 ± 26.2[b]	530.4 ± 47.3[ab]	461.5 ± 29.8[c]
	Cys	187.9 ± 15.5[a]	159.6 ± 8.7[b]	164.6 ± 26.2[ab]	157.7 ± 15.5[b]
	Met	325.9 ± 14.8[a]	290.8 ± 16.9[b]	293.3 ± 31.0[b]	234.9 ± 15.8[c]
	Ile	463.4 ± 10.2[a]	407.0 ± 23.8[b]	416.2 ± 38.7[b]	359.8 ± 21.6[c]
	Leu	773.9 ± 20.3[a]	689.5 ± 41.3[bc]	715.5 ± 64.3[b]	647.4 ± 35.7[c]
	Tyr	315.4 ± 7.7[a]	287.1 ± 13.1[ab]	299.3 ± 36.2[ab]	266.9 ± 21.2[b]
	Phe	481.2 ± 14.1[a]	435.2 ± 30.9[bc]	464.1 ± 41.7[ab]	410.8 ± 22.8[c]
	Lys	645.7 ± 19.7[a]	574.6 ± 36.6[b]	592.6 ± 51.8[b]	523.7 ± 37.4[c]
	His	231.3 ± 9.6[a]	192.0 ± 19.0[b]	207.1 ± 22.5[b]	190.0 ± 13.5[b]
	Arg	553.9 ± 15.3[a]	483.4 ± 34.1[b]	514.5 ± 45.5[ab]	508.6 ± 32.8[ab]
	Pro	334.9 ± 17.2[b]	308.0 ± 20.2[b]	301.2 ± 29.4[b]	386.4 ± 27.7[a]
	Total	8,897.0 ± 269.1[a]	7,960.1 ± 459.9[b]	8,337.8 ± 777.2[ab]	7,805.0 ± 460.2[b]

[a–d]Means in the same row with no common superscript differ ($P < 0.05$).

[1]Values are means ± SD for five eggs and embryos.

[2]Asp = aspartic acid; Thr = threonine; Ser = serine; Glu = glutamic acid; Gly = glycine; Ala = alanine; Val = valine; Cys = cystine; Met = methionine; Ile = isoleucine; Leu = leucine; Tyr = tyrosine; Phe = phenylalanine; Lys = lysine; His = histidine; Arg = arginine; and Pro = proline.

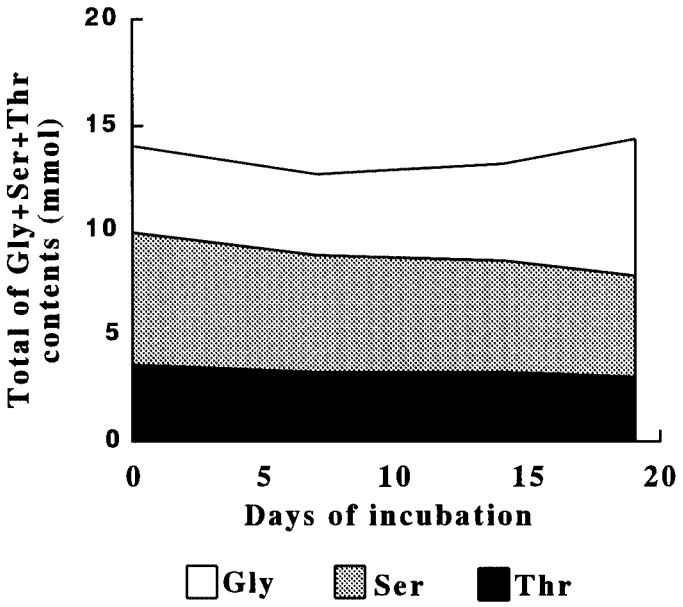

FIGURE 4. Change in total contents of glycine (Gly), serine (Ser), and threonine (Thr) of hatching eggs during incubation.

injection in Experiment 2 (Table 1). Injecting AA into the yolk sac or air cell at Day 0 of incubation resulted in hatchability of 13.3% (Table 5). In the groups treated on Day 7 of incubation, zero hatchability was observed in the group in which the AA solution was injected into the air cell, whereas hatchability of the group injected into the yolk sac was similar to that of the control group at Day 7. There was no significant difference in BW of hatched chicks among all treatments. However, hatching BW relative to egg weight was improved when AA solution was injected into the yolk sac at Day 7 of incubation.

DISCUSSION

In mammals, the nutrition of nursing or postnursing animals affects the subsequent growth and carcass composition of progeny (Knittle and Hirsch, 1968; Winick and Noble, 1967). In poultry, chicks are affected by the nutrients in yolk remaining in the peritoneal cavity posthatching (Romanoff, 1960). Because fat and moisture, but not protein, are in excess (Al-Murrani, 1978), embryonic and postembryonic growth may be improved by AA injection into the egg (Al-Murrani, 1982).

The ratios of transferred CP, CF, and moisture into embryo were 58, 29, and 64% of egg, respectively, on Day 19 of incubation. The lowest transfer ratio for CF might have been due to the fact that CF is used as an energy source by the embryo (Noble, 1986). The AA pattern of egg was constant throughout incubation. The AA of albumen are transferred to the embryo at the same ratio, regardless of incubation time (Rupe and Farmer, 1955). Albumen is absorbed into the yolk sac (Freeman and Vince, 1974), and AA in yolk and albumen may be utilized at a constant ratio. All AA contents of eggs or embryos, except Gly and Pro, changed similarly with advancing incubation time. This relationship suggests that the AA pattern of egg is an ideal pattern for embryonic growth. However, the proportion of Gly and Pro in the embryo increased with incubation time. Because Gly and Pro are used for collagen synthesis, and collagen is required for tissue synthesis, Gly and Pro may be synthesized from other AA during incubation. Proline is synthesized from several AA, whereas precursors of Gly are limited to only Thr and Ser. Total molecular weight of Gly, Ser, and Thr remained constant throughout incubation time (Figure 4). Therefore, it was concluded that the pattern of AA injected in the embryo should be the same as that in the egg prior to incubation.

TABLE 5. Effect of holing, insert of needle, and injection of water or amino acid (AA) mixture on hatching and hatched body weight of broilers[1]

Treated				Initial	Body weight of	Body weights/
Day	Position	Injection	Hatchability[2]	egg weight	hatched chicks	initial egg weight
			(%)	(g)		(%)
		Control	73.3	68.7 ± 4.5	52.0 ± 4.1	75.7 ± 1.8[b]
0	Air cell	None	86.7	66.9 ± 2.7	51.3 ± 2.8	76.7 ± 1.7[ab]
0	Air cell	Water	73.3	66.8 ± 3.5	51.4 ± 3.0	77.0 ± 1.6[ab]
0	Air cell	AA	13.3*	65.2 ± 1.2	49.9 ± 3.0	74.5 ± 2.8[b]
0	Yolk sac	None	66.7	68.0 ± 3.0	51.9 ± 2.6	76.3 ± 1.5[b]
0	Yolk sac	Water	33.3*	67.8 ± 5.7	52.1 ± 4.5	77.0 ± 2.8[ab]
0	Yolk sac	AA	13.3*	65.9 ± 0.4	50.5 ± 1.0	76.2 ± 0.9[b]
7	Air cell	None	86.7	67.8 ± 4.0	52.1 ± 3.8	76.8 ± 2.0[ab]
7	Air cell	Water	86.7	67.9 ± 4.0	51.7 ± 4.0	76.0 ± 2.4[b]
7	Air cell	AA	ND[3]			
7	Yolk sac	None	66.7	67.0 ± 3.0	51.7 ± 3.0	77.1 ± 1.7[ab]
7	Yolk sac	Water	77.3	67.5 ± 3.4	51.2 ± 4.0	75.9 ± 2.7[b]
7	Yolk sac	AA	66.7	68.2 ± 4.1	53.9 ± 3.1	79.0 ± 2.0[a]

[a,b]Means in the same column with no common superscript differ ($P < 0.05$).

[1]Values are means ± SD for 15 eggs.

[2]Hatchability data includes nonfertilized egg because some eggs were treated on Day 0 of incubation.

[3]ND = Not detectable.

*Significantly different from control group ($P < 0.05$).

There are few studies on optimum time and site for injection of AA into egg. Adequate time and position for AA injection into egg was evaluated in Experiment 2. The results obtained suggest that the AA solution injected into egg yolk may be an effective means to increase chick size without decreasing hatchability. Also, better results were obtained when the AA were injected into the egg yolk on Day 7 of incubation. Results varied as AA administration site varied. Embryos absorb nutrients (including free AA) from yolk sac membrane and oxygen from the chorion that exists in the air cell (Romanoff, 1960). Therefore, differences may be caused by the sensitivity of the chorion to AA, whereas the yolk sac membrane is not.

The AA content was higher in the egg than in the embryo at Day 19 of incubation (Table 4), which suggests that the AA content of eggs is sufficient for hatching. Al-Murrani (1982) indicated that about 55 mg AA administered to the egg improved protein accumulation of embryo by about 400 mg. Thus, the effect of AA injection on BW of hatched chicks and egg weight in Experiment 2 might have been due to the increased AA content of yolk or the possibility that AA administration heightened the AA utilization of the embryo.

ACKNOWLEDGMENT

The authors thank Kyowa Hakko Kogyo Co. Ltd. for assistance and AA analysis of the eggs and embryos.

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