

# Effect of the Genetic Selection of Turkeys for Increased Body Weight and Egg Production on Immune and Physiological Responses<sup>1</sup>

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**ABSTRACT** Selection of poultry for fast growth rate is often accompanied by a reduction in specific immune responses or increased disease susceptibility. In this study, 17-wk-old male turkeys from each of four closed genetic lines, a randombred control (RBC) line and its subline (F) selected for increased 16-wk BW, and another RBC line and its subline (E) selected for increased egg production, were tested for *in vivo* response to toe web inoculation with phytohemagglutinin-P (PHA-P), *in vitro* response of lymphocytes in whole blood to PHA-P and concanavalin A (Con A), hemolytic complement activity, differential white blood cell counts, hematology, and serum chemistry values. Fifteen male turkeys from each of two commercial lines, Com A and Com B, were also tested.

The large-bodied F line birds had a lower toe web response to PHA-P, lower lymphocyte counts, and lower

relative spleen weights than their smaller parent line. Body weights, total erythrocyte counts, blood urea nitrogen (BUN) levels, and *in vitro* mitogenic response to PHA-P and Con A were higher in the F line birds. Line E had lower hemolytic complement levels, lower relative spleen and relative bursal weights, and a higher *in vitro* mitogenic response to PHA-P than its parent line. The Com B line had a lower toe web response to PHA-P, and lower serum levels of  $\gamma$ -glutamyltransferase and bilirubin than Com A. Line Com B had higher total RBC counts and higher levels of alanine aminotransferase (ALT) than Com A. These results support the concept that some changes in the cell-mediated immune response, as well as other physiological changes that may potentially affect immune response, appear to accompany selection for faster growth.

(Key words: turkey, body weight, egg production, immune response, blood chemistry)

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## INTRODUCTION

Genetic selection of poultry for superior growth rate may be coincidentally accompanied by decreased resistance to disease or reduced immunological response (Han and Smyth, 1972; Saif *et al.*, 1984; Sacco *et al.*, 1991, 1994a; Miller *et al.*, 1992; Tsai *et al.*, 1992; Qureshi and Havenstein, 1994). The reciprocal situation, selection for improved immunological response, has also been shown to result in decreased BW (Siegel and Gross, 1980; Siegel *et al.*, 1982; van der Zijpp, 1983; Okada *et al.*, 1988; Martin *et al.*, 1990; Afraz *et al.*, 1994). Unintentional selection for reduced immune response in large-bodied

lines may have led to decreased resistance to disease as well as an increase in metabolic defects in pathways dependent on the immune response and its products (Cook, 1994).

Specific defects in the immune response may actually enhance performance, as immune stimulation has been shown to have a negative effect on productivity (Klasing *et al.*, 1987; Chamblee *et al.*, 1992; Cook *et al.*, 1993; Roura *et al.*, 1993). The necessity of maintaining breeding stock in relatively clean and biosecure facilities may also contribute to the selection of birds with relatively low ability to respond to the intensity and diversity of antigenic challenge present in field conditions (Gavora, 1990). This practice will still select for lines with low mortality; however, the ability to resolve chronic or latent infections may be diminished. The purpose of this study was to compare the cellular immune function, hematology, and blood chemistry of 17-wk-old male turkeys selected for increased BW or increased egg production with their randombred parental lines and of two strains of commercial turkeys.

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## MATERIALS AND METHODS

### Turkeys

Thirteen to sixteen male turkeys from each of four closed genetic lines were studied: a randombred control line (RBC1) and its subline (E) selected exclusively for increased egg production over a 180-d period, and another randombred control line (RBC2) and its subline (F) selected for increased 16-wk body weight. These birds were the progeny of a 2-wk hatch of eggs obtained from the Ohio Agricultural Research and Development Center (OARDC) at The Ohio State University. Further information regarding the history of these lines is available in the following references: (McCartney, 1964; McCartney *et al.*, 1968; Nestor, 1977, 1984; Anthony *et al.*, 1991). These turkeys were reared in floor pens, given *ad libitum* access to a standard corn and soybean turkey ration meeting or exceeding the NRC recommended allowances (National Research Council, 1994), and kept on a continuous light schedule. Sixteen and seventeen male turkeys from each of two commercial strains (Com A and Com B, respectively) were obtained from commercial flocks hatched on the same day as the closed lines. The commercial strains were 16 wk of age when received and were held in the same floor pen facility as the closed lines for 1 wk before testing began.

### Bleeding and Necropsy

At 16 wk and 6 d of age all birds were bled by venipuncture into untreated syringes and duplicate smears of blood were made for white blood cell (WBC) differential determinations. Serum was obtained within 2 h of collection. Blood was also collected in heparinized syringes for the whole blood mitogenic assay and hematology determinations. The following day, the birds were weighed, euthanatized using CO<sub>2</sub> asphyxiation, and necropsied. Bursa of Fabricius, liver, and spleen relative weights were determined.

### Cutaneous Basophil Hypersensitivity

Cutaneous basophil hypersensitivity was determined by injecting the toe web with phytohemagglutinin-P (PHA-P) using the methodology of Corrier and DeLoach (1990). The right foot of each bird was cleansed of litter and debris using 70% alcohol. The thickness of the toe web between the third and fourth digits was measured in mm using a micrometer. One hundred microliters of a 1 mg/mL solution of (PHA-P)<sup>4</sup> in sterile 0.85% saline was then

injected intradermally. Twenty-four hours later the toe webs were measured again. Previous studies (data not shown), have shown there to be no differences between the preinjection measurements and postinjection measurements 24 h after a control injection of saline in the left foot of both Com A and Com B turkeys, therefore this control was not used. A stimulation index (SI) was calculated by subtracting the preinjection measurement from the postinjection measurement and dividing the difference by the preinjection measurement.

### In Vitro Mitogenesis

The *in vitro* mitogenic response to PHA-P and Concanavalin A (Con A)<sup>4</sup> was determined using a whole-blood assay based on that reported by Sharma and Belzer (1992). Heparinized whole blood was diluted 1:40 in RPMI-1640 medium<sup>4</sup> containing 2% pooled, heat-inactivated turkey serum (30 min at 56 C), 2 mM L-glutamine, 50 U penicillin, and 50 µg streptomycin per mL. One-hundred and ninety-five microliters of RPMI medium containing the following was added to triplicate wells in each row of a 96-well microtiter plate: 32 µg PHA-P, 3.2 µg PHA-P, and 2 µg Con A. Triplicate wells of RPMI containing no mitogen were included in each row. Five microliters of each diluted blood sample was added to each of the 12 wells of a row. Each plate was placed on a rocking shaker for 10 min, then was incubated at 40 C with 5% CO<sub>2</sub> for 42 to 44 h. One µCi of H<sup>3</sup>-thymidine<sup>5</sup> was added to each well and the plates were incubated for an additional 5 h. The wells were harvested onto glass fiber filters and deposited in vials using a PHD cell harvester.<sup>6</sup> Four milliliters of Scinti-safe Econo-2 liquid scintillation cocktail<sup>7</sup> was added to each vial and radioactivity was measured in CPM using a Beckman model LS6000TA scintillation counter.<sup>8</sup> The mean of the triplicate cultures for each test were determined and divided by the mean proliferative response of unstimulated controls to obtain a SI. A value for the relative degree of mitogenesis per lymphocyte was determined by dividing the SI by the relative number of lymphocytes from each bird as determined by counting the lymphocytes in 25 fields on each of two separate slides.

### Hemolytic Complement Assay

Complement activity was determined in a radial hemolytic plate assay using SRBC sensitized with rabbit hemolysin<sup>4</sup> using the procedure of Skeeles *et al.* (1980). All reagents and buffers were prepared according to Barta and Barta (1975). Hemolytic radial diffusion plates incorporated 0.5 mL of sensitized SRBC per plate. Plates were stored at 4 C until utilized. Wells were cut into the agar using a 4-mm well cutter and a template with four rows of six wells. Agar plugs were removed using vacuum suction. Serum samples were thawed and diluted in twofold steps in NaCl-Veronal buffer with Ca and Mg (Barta and Barta, 1975). Twenty microliters of each serum

<sup>4</sup>Sigma Chemical Co., St. Louis, MO 63178-9916.

<sup>5</sup>Moravek Biochemicals Inc., Brea, CA 92621.

<sup>6</sup>Cambridge Technology, Watertown, MA 02172.

<sup>7</sup>Fisher Scientific, Fair Lawn, NJ 07410.

<sup>8</sup>Model LS6000TA, Beckman Instruments, Inc., Fullerton, CA 92634-3100.

TABLE 1. Body weight, bursa of Fabricius, and spleen relative weights, and the ratio of bursa weight to spleen weight for six turkey lines

Turkey line <sup>1</sup>	n	BW (kg)	Bursa of	Spleen	Bursa:spleen
			Fabricius	(% BW)	ratio
RBC1	16	5.8 ± 0.1	0.08 ± 0.01	0.07 ± 0.01	1.2 ± 0.1
E	13	4.9 ± 0.1****	0.07 ± 0.01*	0.06 ± 0.01*	1.3 ± 0.1
RBC2	14	7.1 ± 0.1	0.09 ± 0.01	0.08 ± 0.01	1.2 ± 0.1
F	15	10.8 ± 0.3****	0.12 ± 0.01	0.04 ± 0.01****	3.3 ± 0.4****
Com A	16	11.5 ± 0.3	0.11 ± 0.01	0.05 ± 0.01	2.1 ± 0.2
Com B	17	11.0 ± 0.2	0.10 ± 0.01	0.05 ± 0.01	2.0 ± 0.2
Small	43	6.0 ± 0.1	0.08 ± 0.01	0.07 ± 0.01	1.2 ± 0.7
Large	48	11.1 ± 0.2****	0.11 ± 0.01	0.05 ± 0.01****	2.5 ± 0.2***

<sup>1</sup>RBC1 = randombred control line; E = subline of RBC1 selected for increased egg production; RBC2 = randombred control line; F = subline of RBC2 selected for increased 16-wk BW; Com A and Com B = commercial turkey lines; Large lines = F, Com A, and Com B; Small lines = RBC1, RBC2, and E. Data are the mean ± SEM.

\* $P \leq 0.05$ .

\*\*\*\* $P \leq 0.0001$ .

dilution from 1:8 through 1:256 was deposited into individual wells. Serum was allowed to diffuse into the agar for 15 min at room temperature. Plates were then incubated at 37 C for 1 h with high humidity. Immediately following incubation, plates were read on a light box. Wells were considered positive if there was a hemolytic ring present. Titers were recorded as the last dilution that gave a positive reaction.

### White Blood Cell Differential Counts

Duplicate smears were prepared from each blood sample using a cover glass technique (Campbell, 1988) and were stained with the Diff Quik Stain Set.<sup>9</sup> Twenty-five fields on each duplicate slide were examined using a light microscope and 1,000× magnification. Relative numbers of heterophils, lymphocytes, monocytes, eosinophils, and basophils were determined.

### Hematology and Clinical Chemistry

An Oximeter Model OSM3 analyzer<sup>10</sup> was used to measure total hemoglobin. Hematocrit was measured in heparinized capillary tubes after centrifugation. Total red blood cell (RBC) counts were made using a Coulter Model ZM counter.<sup>11</sup> Serum concentrations of uric acid, iron, total protein, blood urea nitrogen (BUN), albumin, alkaline phosphatase (AP), alanine aminotransferase (ALT), aspartate aminotransferase (AST),  $\gamma$ -glutamyl-transferase (GGT), and lactate dehydrogenase (LDH) were determined using a Ciba-Corning Express Plus chemistry analyzer and the reagents and procedures standardized for use with the analyzer.<sup>12</sup>

### Statistical Analysis

The *t* test procedure of SAS<sup>®</sup> (SAS Institute, 1988) was used to compare tests between each randombred line and its subline and between each of the two commercial strains. The three strains selected for increased BW (F, Com A, and Com B) were also compared to those not selected for increased BW (RBC1, RBC2, and E). All organ weights were expressed as a percentage of body weight and were subjected to arc sine transformation before analysis.

## RESULTS

Body weight was significantly greater in the RBC1 line than in the E line, greater in the F line than in the RBC2 line, and greater in the large lines than in the small lines (Table 1). There were no differences in relative liver weight (data not shown). Relative bursal weight was greater in the RBC1 line than in the E line and greater in the large lines than in the small lines (Table 1). Relative spleen weight was greater in RBC1 than in the E line, greater in RBC2 than in the F line and greater in the small lines than in the large lines. The ratio of bursa weight to spleen weight was higher in the F line than in RBC1 and greater in the large lines as compared to the small lines.

Toe web hypersensitivity SI was lower in the F line than in its parent line, RBC2, and lower in Com A than in Com B (Table 2). There was no difference between the E line and RBC1 nor in large vs small lines. The total *in vitro* mitogenic response to PHA-P was greater in Line E than in RBC1 before (PHA-P) and after (PHA-P/Lym) correction for lymphocyte numbers whereas response of Line F was less than RBC2 before correction but greater than RBC2 after correction for lymphocyte numbers. Total *in vitro* mitogenic response to Con A was higher in

<sup>9</sup>Baxter Healthcare Corp., Miami, FL 33152-0672.

<sup>10</sup>Radiometer America, Inc., Westlake, OH 44145.

<sup>11</sup>Coulter Corp., Hialeah, FL 33012.

<sup>12</sup>Ciba-Corning Diagnostic Corp., Medfield, MA 02052.

TABLE 2. Responses of turkey lines to toe web inoculation of phytohemagglutinin-P (PHA-P), and *in vitro* stimulation of peripheral blood lymphocytes with PHA-P and Concanavalin A (Con A)

Turkey line <sup>1</sup>	n	Toe web <sup>2</sup>	PHA-P <sup>2</sup>	PHA-P/Lym <sup>3</sup>	Con A <sup>2</sup>	Con A/Lym <sup>3</sup>
RBC1	16	0.33 ± 0.06	1.5 ± 0.2	0.01 ± 0.01	3.9 ± 0.8	0.02 ± 0.01
E	13	0.33 ± 0.07	3.8 ± 1.2*	0.03 ± 0.01*	3.1 ± 0.7	0.01 ± 0.01
RBC2	14	0.34 ± 0.07	0.9 ± 0.1	0.01 ± 0.01	2.4 ± 0.3	0.09 ± 0.01
F	15	0.17 ± 0.04*	0.4 ± 0.5*	0.02 ± 0.01**	7.0 ± 0.9****	0.03 ± 0.01**
Com A	16	0.48 ± 0.03	2.6 ± 0.7	0.03 ± 0.01	4.5 ± 0.9	0.02 ± 0.01
Com B	17	0.29 ± 0.04**	2.9 ± 0.8	0.04 ± 0.01	2.4 ± 0.4*	0.02 ± 0.01
Small	43	0.34 ± 0.04	2.0 ± 0.4	0.02 ± 0.01	3.1 ± 0.4	0.01 ± 0.01
Large	48	0.32 ± 0.03	2.7 ± 0.4	0.03 ± 0.01	4.6 ± 0.5*	0.02 ± 0.01*

<sup>1</sup>RBC1 = randombred control line; E = subline of RBC1 selected for increased egg production; RBC2 = randombred control line; F = subline of RBC2 selected for increased 16-wk BW; Com A and Com B = commercial turkey lines; Large lines = F, Com A, and Com B; Small lines = RBC1, RBC2, and E.

<sup>2</sup>Data are the mean ± SEM of the stimulation index (SI).

<sup>3</sup>Data are the mean ± SEM of the SI divided by the total lymphocyte count in 50 fields at 1,000.

\* $P \leq 0.05$ .

\*\* $P \leq 0.01$ .

\*\*\*\* $P \leq 0.0001$ .

the F line than in RBC2 before and after correction for lymphocyte numbers, higher in Com A than in Com B without correction for lymphocyte numbers, and higher in large lines than in small lines for both corrected and uncorrected data.

The number of lymphocytes was significantly lower in the F line than in RBC2, and lower in large lines than in small lines (Table 3). There was a significant increase in heterophil numbers and in the heterophil to lymphocyte ratio in large lines than in small lines (Table 3). The large lines also had a significantly higher number of monocytes and a higher total count than in the small lines (Table 3). There were no differences in eosinophil or basophil counts (data not shown).

Hemolytic complement levels and hemoglobin levels were higher in the RBC1 line than in the E line (Table 4).

Total erythrocyte counts were higher in the E Line than in RBC1, higher in the F line than in RBC2, higher in Com B than in Com A, and higher in large lines than in small lines. Hematocrit was higher in Com B as compared to Com A and iron levels were higher in large lines as compared to small lines.

The Com B line had higher serum levels of ALT and lower levels of GGT (Table 5) than Com A. Large lines had higher levels of ALT and AST and lower levels of AP than small lines. There were no differences in levels of LDH (data not shown).

Large lines had higher levels of total protein than small lines (Table 6). Blood urea nitrogen levels were higher in the F line than in RBC2, higher in Com B than in Com A, and higher in large lines than in small lines.

TABLE 3. Differential white blood cell counts<sup>1,2</sup> of turkey lines

Turkey line <sup>3</sup>	n	L	H	H:L ratio	M	Total
RBC1	16	112 ± 8	76 ± 6	0.70 ± 0.05	9 ± 1	201 ± 13
E	13	116 ± 10	80 ± 5	0.74 ± 0.05	12 ± 2	214 ± 15
RBC2	14	132 ± 4	91 ± 9	0.70 ± 0.05	17 ± 3	246 ± 19
F	15	100 ± 8*	110 ± 25	1.25 ± 0.39	25 ± 6	241 ± 30
Com A	16	100 ± 7	183 ± 28	1.97 ± 0.35	28 ± 4	317 ± 31
Com B	17	102 ± 8	132 ± 22	1.39 ± 0.24	33 ± 7	274 ± 31
Small	43	120 ± 6	82 ± 4	0.71 ± 0.03	12 ± 1	19 ± 9
Large	48	101 ± 4*	142 ± 15***	1.53 ± 0.19****	28 ± 3****	277 ± 18

<sup>1</sup>Data are the mean ± SEM of the numbers of each cell type counted in 50 fields on two replicate slides using 1,000× magnification.

<sup>2</sup>L = lymphocyte; H = heterophil; H/L = heterophil:lymphocyte ratio; M = monocyte.

<sup>3</sup>RBC1 = randombred control line; E = subline of RBC1 selected for increased egg production; RBC2 = randombred control line; F = subline of RBC2 selected for increased 16-wk BW; Com A and Com B = commercial turkey lines; Large lines = F, Com A, and Com B; Small lines = RBC1, RBC2, and E.

\* $P \leq 0.05$ .

\*\*\* $P \leq 0.001$ .

\*\*\*\* $P \leq 0.0001$ .

TABLE 4. Hemolytic complement levels and hematology values for turkey lines

Turkey line <sup>1</sup>	n	Hemolytic complement (-Log titer)	Total erythrocytes ( $\times 10^6/\text{mL}$ )	Hematocrit (%)	Hemoglobin (g/100 mL)	Iron ( $\mu\text{g}/\text{dL}$ )
RBC1	16	1.43 $\pm$ 0.08	2.3 $\pm$ 0.1	36.4 $\pm$ 0.7	12.0 $\pm$ 0.2	196 $\pm$ 32
E	13	1.18 $\pm$ 0.04*	3.2 $\pm$ 0.1****	34.9 $\pm$ 0.6	11.5 $\pm$ 0.1*	178 $\pm$ 38
RBC2	14	0.58 $\pm$ 0.51	2.4 $\pm$ 0.1	36.9 $\pm$ 0.7	12.2 $\pm$ 0.2	150 $\pm$ 30
F	15	1.30 $\pm$ 0.07	3.1 $\pm$ 0.1	37.0 $\pm$ 0.6	11.9 $\pm$ 0.4	112 $\pm$ 27
Com A	16	1.32 $\pm$ 0.07	2.4 $\pm$ 0.1	35.5 $\pm$ 0.5	11.7 $\pm$ 0.3	129 $\pm$ 20
Com B	17	0.89 $\pm$ 0.43	3.1 $\pm$ 0.1****	37.4 $\pm$ 0.8*	12.1 $\pm$ 0.4	91 $\pm$ 12
Small	43	1.08 $\pm$ 0.17	2.6 $\pm$ 0.1	36.1 $\pm$ 0.4	11.9 $\pm$ 0.1	175 $\pm$ 19
Large	48	1.16 $\pm$ 0.16	2.8 $\pm$ 0.1*	36.6 $\pm$ 0.4	11.9 $\pm$ 0.2	110 $\pm$ 11*

<sup>1</sup>RBC1 = randombred control line; E = subline of RBC1 selected for increased egg production; RBC2 = randombred control line; F = subline of RBC2 selected for increased 16-wk BW; Com A and Com B = commercial turkey lines; Large lines = F, Com A, and Com B; Small lines = RBC1, RBC2, and E. Data are the mean  $\pm$  SEM.

\* $P \leq 0.05$ .

\*\*\*\* $P \leq 0.0001$ .

Large lines also had higher levels of bilirubin than small lines, whereas Com A had higher levels of bilirubin than Com B. There were no differences in levels of albumin or uric acid, or in the total protein:albumin ratios (data not shown).

## DISCUSSION

In this study, both lymphocyte numbers and the hypersensitivity reaction to PHA-P were significantly decreased in the F line, which was selected for increased BW, but not in the line selected for increased egg production. The mean relative spleen weight of the F line birds was half that of its parent line, and was significantly lower in large lines than in small lines. Also, Com B, which has a faster early growth rate than Com A (data not shown), had a lower toe web response. However, the *in vitro* assays for T cell function showed

that both PHA-P and Con A induced significantly higher mitogenic activity in the line selected for higher BW, although no differences were seen between Com A and Com B. Although these differences in mitogenic SI were significant, it should be noted that they were relatively low values, and that the low response may be related to the age of the birds. Sharma and Belzer (1992), in a study of the effect of age on mitogenic response to Con A, found that at 6 and 7 wk, and again at 11 and 12 wk of age, the response decreased to the very low levels seen at 1 wk of age.

The lack of correlation between mitogenic activity and the toe-web response has been reported before (Bacon and Heller, 1989) and may reflect differences in the specific T cell populations measured by each test. Previous studies have also shown a lack of correlation between the degree of graft-vs-host response (GVHR) and response to PHA injection (Lamont and Smyth,

TABLE 5. Serum levels of alanine aminotransferase (ALT),  $\gamma$ -glutamyltransferase (GGT), alkaline phosphatase (AP), and aspartate aminotransferase (AST) for the turkey lines

Turkey line <sup>1</sup>	n	ALT	GGT	AP	AST
(U/L)					
RBC1	16	7.8 $\pm$ 0.9	1.4 $\pm$ 0.2	1768 $\pm$ 217	279 $\pm$ 28
E	13	5.9 $\pm$ 0.6	1.0 $\pm$ 0.6	1662 $\pm$ 87	291 $\pm$ 11
RBC2	14	9.9 $\pm$ 0.5	1.5 $\pm$ 0.2	1611 $\pm$ 105	366 $\pm$ 24
F	15	11.1 $\pm$ 1.1	0.8 $\pm$ 0.4	1477 $\pm$ 123	419 $\pm$ 15
Com A	16	7.2 $\pm$ 1.1	3.9 $\pm$ 1.2	1250 $\pm$ 78	413 $\pm$ 41
Com B	17	14.0 $\pm$ 2.0*	1.2 $\pm$ 0.2****	1080 $\pm$ 112	370 $\pm$ 27
Small	43	8.1 $\pm$ 0.5	1.3 $\pm$ 0.2	1685 $\pm$ 93	14 $\pm$ 16
Large	48	10.8 $\pm$ 1.0*	2.3 $\pm$ 0.6	1230 $\pm$ 63****	398 $\pm$ 19***

<sup>1</sup>RBC1 = randombred control line; E = subline of RBC1 selected for increased egg production; RBC2 = randombred control line; F = subline of RBC2 selected for increased 16-wk BW; Com A and Com B = commercial turkey lines; Large lines = F, Com A, and Com B; Small lines = RBC1, RBC2, and E. Data are the mean  $\pm$  SEM.

\* $P \leq 0.05$ .

\*\*\* $P \leq 0.001$ .

\*\*\*\* $P \leq 0.0001$ .

TABLE 6. Serum chemistry values of turkey lines

Turkey line <sup>1</sup>	n	Total protein	Blood urea nitrogen	Bilirubin
		(g/dL)	(mg/dL)	(mg/dL)
RBC1	15	3.2 ± 0.4	1.15 ± 0.09	0.04 ± 0.02
E	9	3.5 ± 0.1	1.20 ± 0.06	0.01 ± 0.01
RBC2	14	3.5 ± 0.2	0.88 ± 0.05	0.02 ± 0.00
F	15	3.9 ± 0.4	1.22 ± 0.12*	0.02 ± 0.01
Com A	16	3.6 ± 0.3	0.94 ± 0.07	0.09 ± 0.01
Com B	17	3.9 ± 0.1	1.54 ± 0.10****	0.05 ± 0.01*
Small	37	3.3 ± 0.2	1.06 ± 0.05	0.02 ± 0.01
Large	41	3.8 ± 0.2*	1.24 ± 0.07*	0.06 ± 0.01*

<sup>1</sup>RBC1 = randombred control line; E = subline of RBC1 selected for increased egg production; RBC2 = randombred control line; F = subline of RBC2 selected for increased 16-wk BW; Com A and Com B = commercial turkey lines; Large lines = F, Com A, and Com B; Small lines = RBC1, RBC2, and E. Data are the mean ± SEM.

\* $P \leq 0.05$ .

\*\*\*\* $P \leq 0.0001$ .

1984), and between delayed wattle response to *Bacillus Calmette-guerin* (BCG) injection, GVHR, and response to PHA injection (Afraz *et al.*, 1994), which suggests that these tests may all measure different aspects of cell-mediated immunity (Afraz *et al.*, 1994).

The decrease seen here in relative spleen weights of fast-growing birds may suggest that a difference in some splenic cell population may be involved in differences seen in T cell response. Kromer *et al.* (1985) reported that obese strain chickens were deficient in a suppressor serum factor that interfered with interleukin-2 function in splenocyte cultures, and that was also associated with Con A hyperresponsiveness. The spleen is an essential lymphoid organ, which has a prominent role in the development of suppressor T cells and which specifically affects distinct populations of lymphoid cells in other tissues (Welles and Battisto, 1978). The increased bursa:spleen ratio of the F line and the large lines may indicate a subtle redistribution of lymphoid tissue in fast-growing birds that may be reflected in the immune response.

The T cell-mediated immune response of chickens has significant variation among birds of different genetic lineages (Miggiano *et al.*, 1976; Lassila *et al.*, 1979; Morrow and Abplanalp, 1981; Fredericksen and Gilmour, 1983; Lamont and Smyth, 1984; Cheng and Lamont, 1988). Successful divergent selection of chickens for various T cell functions, suggests that many of these functions are highly heritable, and are often negatively correlated with BW (Sengar *et al.*, 1970; Okada and Mikami, 1974; Yamamoto and Okada, 1990; Afraz *et al.*, 1994).

The heritability of cell-mediated immune function in the turkey has not been reported; however, considerable data has been generated describing the humoral immune response of the OARDC turkey lines that are described in this study (Sacco *et al.*, 1994a,b). The F line had higher mortality due to natural outbreaks of erysipelas and fowl cholera than its parent line, RBC2,

and the E line had higher mortality than its parent line in an outbreak of fowl cholera (Saif *et al.*, 1984). The E line had lower antibody responses to both Newcastle disease virus (NDV) and *Pasteurella multocida* than did its parent line, RBC1 (Sharaf *et al.*, 1988a) and had higher mortality in one of three challenges with the same organism (Sharaf *et al.*, 1988b). All four lines were studied in separate *P. multocida* (Sacco *et al.*, 1991) and NDV challenges (Tsai *et al.*, 1992). The F line had earlier clinical signs and mortality than the other three lines in the *P. multocida* challenge and had the highest mortality in both challenges. Line F had higher antibody titers to NDV at 9 and 15 wk after vaccination than did RBC2, but had lower antibody titers to *P. multocida* at 15 wk (Sacco *et al.*, 1994b). These studies implicate selection for increased BW, and to a lesser extent, selection for egg production, as being correlated with changes in the humoral immune system and susceptibility to disease. The additional data provided by this study extend those differences to parameters conventionally associated with the cellular response.

Significant differences were seen in the serum chemistry profiles and the heterophil:lymphocyte ratios of the fast-growing strains as relative to the slower growing strains. An increased heterophil:lymphocyte ratio is an accepted indicator of stress in the chicken (Davison *et al.*, 1983; Gross and Siegel, 1983; Maxwell, 1993), and could be related to the physiological stress of rapid growth. Differences in liver enzyme levels are hard to explain. These changes may reflect the close association between the immune system and other aspects of physiology, and may have relevance in the study of ascites, tibial dyschondroplasia, and other diseases thought to be associated with fast-growing poultry.

This study has shown that selection for faster growth rate, in both the F-line birds and in Com B, can result in a decrease in the toe web response to PHA-P. The association of this decreased response, together with the decreased antibody response and increased susceptibil-

ity to bacterial infection shown in previous studies, suggests that selection for increased BW of turkeys is accompanied by decreased immunological response.

## REFERENCES

- Afraz, F., Y. Yamamoto, and I. Okada, 1994. Divergent selection for delayed-type wattle reaction of domestic fowls to BCG antigen. *Br. Poult. Sci.* 35:47-58.
- Anthony, N. B., D. A. Emmerson, and K. E. Nestor, 1991. Genetics of growth and reproduction in the turkey. 12. Results of long-term selection for increased 180-day egg production. *Poultry Sci.* 70:1314-1322.
- Bacon, L. D., and D. Heller, 1989. The role of cell-mediated immunity in genetic resistance to disease in chickens. Page 8 *in*: Final Report. BARD Project No. US-829-884. USDA, National Agricultural Library, Washington, DC.
- Barta, O., and V. Barta, 1975. Chicken (*Gallus gallus*) hemolytic complement: Optimal conditions for its titration. *Immunol. Commun.* 4:337-351.
- Campbell, T. W., 1988. *Avian Hematology*. Iowa State University Press, Ames, IA.
- Chamblee, T. N., J. R. Thompson, and J. P. Thaxton, 1992. Effect of day old vaccination on broiler performance. *Poultry Sci.* 71 (Suppl. 1):144. (Abstr.)
- Cheng, S., and S. J. Lamont, 1988. Genetic analysis of immunocompetence measures in a white leghorn chicken line. *Poultry Sci.* 67:989-995.
- Cook, M. E., 1994. Nutrition and immunity in poultry. Pages 1-9 *in*: Proceedings of the California Animal Nutrition Conference, Fresno, CA.
- Cook, M. E., C. C. Miller, Y. Park, and M. Pariza, 1993. Immune Modulation by altered nutrient metabolism: Nutritional control of immune-induced growth depression. *Poultry Sci.* 72:1301-1305.
- Corrier, D. E., and J. R. Deloach, 1990. Evaluation of cell-mediated cutaneous basophil hypersensitivity in young chickens by an interdigital skin test. *Poultry Sci.* 69:403-408.
- Davison, T. F., J. G. Rowell, and J. Rea, 1983. Effects of dietary corticosterone on peripheral blood lymphocyte and granulocyte populations in immature domestic fowl. *Res. Vet. Sci.* 34:236-239.
- Fredericksen, T. L., and D. G. Gilmour, 1983. Ontogeny of Con A and PHA responses of chicken blood cells in MHC-compatible lines 6<sub>3</sub> and 7<sub>2</sub>. *J. Immunol.* 130:2528-2533.
- Gross, W. B., and H. S. Siegel, 1983. Evaluation of the heterophil/lymphocyte ratio as a measure of stress in chickens. *Avian Dis.* 27:972-979.
- Gavora, J. S., 1990. Disease genetics. Pages 805-846 *in*: *Poultry Breeding and Genetics*. R. D. Crawford, ed., Elsevier, Amsterdam, The Netherlands.
- Han, P.F.S., and J. R. Smyth, 1972. The influence of growth rate on the development of Marek's disease in chickens. *Poultry Sci.* 51:975-985.
- Klasing, K. C., D. E. Laurin, T. K. Peng, and D. M. Fry, 1987. Immunologically mediated growth depression in chicks: Influence of feed intake, corticosterone and interleukin-1. *J. Nutr.* 117:1629-1637.
- Kromer, G., K. Schauenstein, N. Neu, K. Stricker, and G. Wick, 1985. In vitro T cell hyperreactivity in obese strain (OS) chickens is due to a defect in nonspecific suppressor mechanism(s). *J. Immunol.* 135:2458-2463.
- Lamont, S. J., and J. R. Smyth, Jr., 1984. Effect of selection for delayed amelanosis on immune response in chickens. 2. Cell-mediated immunity. *Poultry Sci.* 63:440-442.
- Lassila, O., T. Nurmi, and J. Eskola, 1979. Genetic differences in the mitogenic response of peripheral blood lymphocytes in the chicken. *J. Immunogenet.* 6:37-43.
- Martin, A., E. A. Dunnington, W. B. Gross, W. E. Briles, R. W. Briles, and P. B. Siegel, 1990. Production traits and alloantigen system in lines of chickens selected for high and low antibody response to sheep erythrocytes. *Poultry Sci.* 69:871-878.
- Maxwell, M. H., 1993. Avian blood leukocyte responses to stress. *World's Poult. Sci. J.* 49:34-43.
- McCartney, M. G., 1964. A randombred control population of turkeys. *Poultry Sci.* 43:739-744.
- McCartney, M. G., K. E. Nestor, and W. R. Harvey, 1968. Genetics of growth and reproduction in the turkey. 2. Selection for increased body weight and egg production. *Poultry Sci.* 47:981-990.
- Miggiano, V., M. North, A. Buder, and J.R.L. Pink, 1976. Genetic control of the response of chicken leukocytes to a T-cell mitogen. *Nature* 263:61-63.
- Miller, C. C., M. E. Cook, G. E. Rodgers, and H. Kohl, 1992. Immune response differences in different strains of ducks. *Poultry Sci.* 71 (Suppl. 1):166. (Abstr.)
- Morrow, P. R., and H. Abplanalp, 1981. Genetic control of T-lymphocyte mitogenesis in chickens. *Immunogenetics* 13:189-200.
- National Research Council, 1994. *Nutrient Requirements of Poultry*. 9th rev. ed. National Academy Press, Washington, DC.
- Nestor, K. E., 1977. The stability of two randombred control populations of turkeys. *Poultry Sci.* 56:54-57.
- Nestor, K. E., 1984. Genetics of growth and reproduction in the turkey. 9. Long-term selection for increased 16-week body weight. *Poultry Sci.* 63:2114-2122.
- Okada, I., H. Bansho, M. Yamamoto, T. Kaizuka, and Y. Yamamoto, 1988. Two-way selection of chicken for antibody titers to *Leucocytozoon caulleryi* under the condition of natural infection. *Jpn. Poult. Sci.* 25:366-374.
- Okado, I., and M. Mikami, 1974. Three generations of selection for high and low donor competencies of splenomegaly in chickens. *Br. Poult. Sci.* 15:1-10.
- Qureshi, M., and G. B. Havenstein, 1994. A comparison of the immune performance of a 1991 commercial broiler with a 1957 randombred strain when fed "typical" 1957 and 1991 broiler diets. *Poultry Sci.* 73:1805-1812.
- Roura, E., and K. C. Klasing, 1993. Dietary antibiotics reduce immunological stress elicited by poor sanitation or consumption of excreta in broiler chicks. *Poultry Sci.* 72 (Suppl. 1):1. (Abstr.)
- Sacco, R. E., K. E. Nestor, Y. M. Saif, H. J. Tsai, N. B. Anthony, and R. A. Patterson, 1994a. Genetic analysis of antibody response of turkeys to Newcastle disease virus and *Pasteurella multocida* vaccines. *Poultry Sci.* 73:1169-1174.
- Sacco, R. E., K. E. Nestor, Y. M. Saif, H. J. Tsai, and R. A. Patterson, 1994b. Effect of genetic selection for increased body weight and sex of poul on antibody response of turkeys to Newcastle disease virus and *Pasteurella multocida* vaccines. *Avian Dis.* 38:33-36.
- Sacco, R. E., Y. M. Saif, K. E. Nestor, N. B. Anthony, D. A. Emmerson, and R. N. Dearth, 1991. Genetic variation in resistance of turkeys to experimental challenge with *Pasteurella multocida*. *Avian Dis.* 35:950-954.

- Saif, Y. M., K. E. Nestor, R. N. Dearth, and P. A. Renner, 1984. Case Report: Possible genetic variation in resistance of turkeys to Erysipelas and fowl cholera. *Avian Dis.* 28: 770-773.
- SAS Institute, 1988. SAS/STAT® User's Guide: 1988 Edition. SAS Institute Inc., Cary, NC.
- Sengar, D.P.S., F. N. Jerome, and B. S. Reinhart, 1970. An attempt to improve tolerance in chickens by selection. *Poultry Sci.* 49:1030-1037.
- Sharaf, M. M., K. E. Nestor, Y. M. Saif, R. E. Sacco, and G. B. Havenstein, 1988a. Antibody response to Newcastle disease virus and *Pasteurella multocida* of two strains of turkeys. *Poultry Sci.* 67:1372-1377.
- Sharaf, M. M., K. E. Nestor, Y. M. Saif, R. E. Sacco, and G. B. Havenstein, 1988b. Research note: Response of two lines of turkeys to challenge with *Pasteurella multocida*. *Poultry Sci.* 67:1807-1809.
- Sharma, J. M., and S. W. Belzer, 1992. Blastogenic response of whole-blood cells of turkeys to a T-cell mitogen. *Dev. Comp. Immunol.* 16:77-84.
- Siegel, P. B., and W. B. Gross, 1980. Production and persistence of antibodies in chickens to sheep erythrocytes. 1. Directional Selection. *Poultry Sci.* 59:1-5.
- Siegel, P. B., W. B. Gross, and J. A. Cherry, 1982. Correlated responses of chickens to selection for production of antibodies to sheep erythrocytes. *Anim. Blood Groups Biochem. Genet.* 13:291-297.
- Skeeles, J. K., R. G. Stewart, J. Brown, R. K. Page, and I. D. Russell, 1980. Hemolytic complement activity in broiler chickens and turkeys. *Poultry Sci.* 59:1221-1225.
- Tsai, H. J., Y. M. Saif, K. E. Nestor, D. A. Emmerson, and R. A. Patterson, 1992. Genetic variation in resistance of turkeys to experimental infection with Newcastle disease virus. *Avian Dis.* 36:561-565.
- Welles, W. L., and J. R. Battisto, 1978. The function of the spleen in immune responses and its influence on other lymphoid organs. Pages 236-249 *in: Animal Models of Comparative and Developmental Aspects of Immunity and Disease.* M. E. Gershwin and E. L. Cooper, ed. Pergamon Press, New York, NY.
- Yamamoto, Y., and I. Okada, 1990. Two-way selection for survival time of allografts in chickens. *Jpn. Poult. Sci.* 27: 337-345.
- van der Zijpp, A. J., 1983. Breeding for immune responsiveness and disease resistance. *World's Poult. Sci. J.* 39:118-131.