

Estimation of crossbreeding effects for growth and immunological traits in a crossbreeding experiment involving two local strains of chickens

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Abstract

Two native strains namely Matrouh (MA) and Inshas (IN) were used in a crossbreeding experiment. A total number of 480 chicks (produced from four genetic groups, two purebred strains and their reciprocal crosses) were chosen randomly to study crossbreeding effects (direct additive, maternal additive and heterosis) for growth traits and immunological trait against *Salmonella*. The traits studied were body weight (BW) at 1st, 2nd, 3rd, 4th, 5th, 6th, 7th, 8th, 9th and 10th week of age; daily gain (DG) during the intervals 1-4, 4-8, 8-10 and 1-10 weeks of age; Salmonella count; caecal pH and antibody titer.

Results showed that: MA strain had superiority in growth traits ($P < 0.05$) and antibody titer compared to IN strain, but lower Salmonella count and caecal pH. Estimates of direct additive genes showed positive effect for growth traits ($P \leq 0.01$) (ranged from 2.22% to 10.4%), Salmonella count ($P \leq 0.05$) (12.1%) and antibody titer (6.32%), but negative for only caecal pH (-1.11%). Estimates of maternal effects were negative for most of the traits studied. There was a highly significant decrease the salmonella count by 18.2%, thus dams of MA strain could be favored to decrease the Salmonella count in Egyptian broiler chickens. Moreover, maternal effect showed a negative on antibody titers at 4th week of age. It was decreased the immunity of chicks against *Salmonella typhimurium* by 7.69%. Estimates of heterosis were negative for most of body weights (ranged from -2.98 to 9.05%) and Salmonella count ($P \leq 0.01$) (-21.2%), but positive for daily gains (ranged from 0.5 to 11.3%) and antibody titer ($P \leq 0.01$) (25.2%).

In conclusion, IN strain could be used as a sire and MA strain as a dam to improve growth and immunological traits. Thus, the birds' immunity against Salmonella could be improved by crossing.

Key words: direct additive and maternal effect, growth traits, heterosis, microbial and immunological traits

Introduction

Breeding for disease resistance in poultry has a long history. Resistance of chickens was selected naturally at the beginning of commercial stocks when no protected areas were available. However, the efficiency of such method was limited as pathogens varied and the lack of knowledge did not always make it possible to distinguish between unrelated diseases (Payne 1973). Avian infectious diseases are costly for the poultry industry. They increase management costs, lead to production losses and raise humane concerns. Avian infectious diseases are also a concern for humans, since chickens may be carriers for pathogens that affect human health. Bacterial pathogens such as *Salmonella* or *Escherichia coli* may cause ill health in humans (Campitelli et al 2002).

Nowadays, more workers on Egyptian native breeds are needed. The Egyptian chickens have the advantages of being well adapted to the local stressful conditions. Another advantage claimed by Egyptian consumers is the good taste and flavor of local chickens' meats and eggs (Kolstad and Abdou 2000). Some studies (Shebl et al 1990 and Iraqi et al 2002&2005) reported that most of the native breeds had high non-additive genetic variance and, therefore, possibility of improvement of these breeds through crossbreeding is evident. Estimating crossbreeding components in a crossbreeding experiment makes it possible to know which strain could be used as sires or dams. There is no available information for crossbreeding effects on bacterial pathogens (e.g. Salmonella count), pH and antibody titer traits in broilers of chickens.

This work was designed to study the effect of genetic groups on growth traits, controlling *Salmonella* colonization in intestinal tract of chickens, pH and immunity, as well as to estimate crossbreeding components (direct additive, maternal effect and heterosis) for traits studied in two local strains of chickens named Matrouh and Inshas and their reciprocal crosses.

Materials and methods

This experiment was carried out in the Poultry Farm of chickens, Faculty of Agriculture at Moshtohor, Benha University, Egypt, started in April 2005 and terminated in June 2005.

Breeding plan and management

Two local strains named Matrouh (MA) and Inshas (IN) were used. Pullets of each of the two strains were randomly divided into two groups (100 hens / group); the first group was mated with 10 cocks from the same strain, while the second group was mated with 10 cocks from the other strain. Consequently, the pedigreed eggs from each individual breeding pen for the four mating groups (Table 1) were collected daily for ten days and incubated in one hatch then after.

Table 1. Number of chicks used in the experimental work and description of genetic group of sires and dams produced from them

Genetic group* of chicks	No. of chicks	Genetic group of sire	Genetic group of dam
MA x MA	120	MA	MA
IN x IN	120	IN	IN
MA x IN	120	MA	IN
IN x MA	120	IN	MA
Total	480		

*MA and IN= Matrouh and Inshas strains, respectively

On hatching day, numbers of 120 chicks (12 chicks from each sire) were randomly chosen from each genetic group, then after wing banded to save its genetic groups and immediately transferred to the Poultry Farm of chickens in April 2005. Chicks from each genetic group were distributed randomly on ten treatments (12 chicks in each). Description of treatments supplied to the chicks is summarized in Table 2.

Table 2. Description of treatments used in the experimental work

Treatment No.	Description of treatment
1	2.5% lactose added in drinking water
2	2.5% lactose in drinking water and <i>Lactobacillus acidophilus</i>
3	2.5% lactose in drinking water and <i>Enterococcus faecalis</i>
4	2.5% lactose in drinking water and <i>Bacillus subtilis</i>
5	2.5% lactose in drinking water and <i>Lactobacillus acidophilus</i> , <i>Enterococcus faecalis</i> , and <i>Bacillus subtilis</i>
6	Control negative group without any treatment
7	Control positive group treated with <i>Salmonella typhimurium</i> only
8	Treated with <i>Lactobacillus acidophilus</i>
9	Treated with <i>Enterococcus faecalis</i>
10	Treated with <i>Bacillus subtilis</i>

At hatch, chicks were challenged with 10^6 (*Lactobacillus acidophilus*, *Enterococcus faecalis*, and *Bacillus subtilis*) by crop inoculation. After three days from hatch, all chicks were challenged with 10^6 *Salmonella typhimurium* by crop inoculation, except the control negative group. Chicks were reared in floor brooder up to end of the experiment under continuous lighting program (fluorescent lamps, 10 watt/m²). They were fed (without antibiotics, coccidiostats, or growth promoters) during rearing and growing periods on diet containing 23.01 %, 20 % crude protein, 3.6 %, 3.19 % crude fiber,

respectively, as well as *ad libitum* drinking water. All birds were subjected to similar hygienic and environmental conditions and vaccinated against Newcastle and Gambaro diseases.

Data and traits studied

Data of 480 chicks were recorded for traits of body weight (g) at 1st (BW1), 2nd (BW2), 3rd (BW3), 4th (BW4), 5th (BW5), 6th (BW6), 7th (BW7), 8th (BW8), 9th (BW9) and 10th (BW10) weeks of age. Daily weight gains during the periods from 1st to 4th, 4th to 8th, 8th to 10th and 1st to 10th weeks of age were computed. Salmonella count and caecal pH traits were also studied, as well as antibody titer in serum was estimated according to procedure of (Alton et al 1988). The antibody titer was expressed as (-log₂).

Statistical analysis

Data of body weight and daily gain traits were analyzed using (Model 1), but Salmonella count, antibody titer and caecal pH traits were analyzed using (Model 2) according to SAS program (SAS 1996).

$$Y_{ijkl} = \mu + G_i + T_j + X_k + (GT)_{ij} + e_{ijkl} \quad (\text{Model 1})$$

$$Y_{ijk} = \mu + G_i + T_j + (GT)_{ij} + e_{ijk} \quad (\text{Model 2})$$

Where:

Y_{ijkl} and Y_{ijk} = the observation recorded on chick;

μ = the overall mean;

G_i = fixed effect of the i^{th} genetic group;

T_j = fixed effect of the j^{th} treatment;

X_k = fixed effect of k^{th} sex (levels= 1, 2 and 3 for males, females and dead chicks before sexing, respectively);

$(GT)_{ij}$ = Fixed effect of interaction between the i^{th} genetic group and j^{th} treatment; and

e_{ijkl} and e_{ijk} = the random deviation particular to the chick, assumed to be independently randomly distributed with zero mean and variance (σ_e^2).

Main effects of genetic group and treatment included in the two models were significant. Effect of the interaction between two main effects was also significant, but the main effects are averaged over the interaction. Duncan's multiple range test (Duncan 1955) was used to detect the significant differences between means of genetic groups.

Estimation of crossbreeding components

Estimates of crossbreeding effects (direct additive, maternal, and heterosis) for all the traits studied were calculated using CBE package (Wolf 1996). Estimates of each component were calculated according to (Dickerson 1992). The following symbols were described in Table 1.

Direct additive effect (G^I):

$$\frac{1}{2} [(MA \times MA - IN \times IN) - (MA \times IN - IN \times MA)]$$

Maternal effect (G^M):

$$\frac{1}{2} [(MA \times IN) - (IN \times MA)]$$

Direct heterosis (H^I):

$$\frac{1}{2} [(IN \times MA + MA \times IN) - (MA \times MA + IN \times IN)]$$

Results and discussion

The means presented in Table 3 indicate that Matrouh (MA) strain was significantly heavier in body weight than Inshas (IN) strain for most of the traits studied, as well as higher in daily gains during the intervals 1-4, 4-8, 8-10 and 1-10 weeks of age.

Table 3. Least-squares means and standard errors for body weight (g), daily gain (g), microbiological and immunological traits as affected by genetic group of chicks

Trait	Genetic group of chicks			
	MA x MA	IN x IN	MA x IN	IN x MA
<i>Body weight (BW)</i>				
BW1	68.8 ^a ±1.0	68.5 ^a ±0.9	65.2 ^b ±1.0	67.9 ^a ±0.9
BW2	104 ^a ±1.6	100 ^{ab} ±1.5	96.2 ^b ±1.6	102 ^a ±1.5
BW3	151 ^a ±2.6	154 ^a ±2.4	136 ^b ±2.5	153 ^a ±2.5
BW4	200 ^a ±3.5	192 ^b ±3.3	187 ^b ±3.4	202 ^a ±3.3
BW5	247 ^a ±9	227 ^b ±8	252 ^a ±9	260 ^a ±9
BW6	324 ^a ±15	279 ^b ±15	320 ^a ±16	331 ^a ±15
BW7	389 ^a ±18	330 ^b ±17	392 ^a ±19	393 ^a ±18
BW8	471 ^a ±21	409 ^b ±21	470 ^a ±22	477 ^a ±22
BW9	607 ^a ±33	526 ^b ±34	609 ^a ±34	603 ^a ±34
BW10	744 ^a ±37	630 ^c ±39	734 ^{ab} ±39	735 ^b ±39
<i>Daily gain (DG)</i>				
DG 1-4	6.26 ^a ±0.14	5.84 ^b ±0.13	5.78 ^b ±0.14	6.38 ^a ±0.14
DG 4-8	9.91 ^a ±0.58	7.90 ^b ±0.57	10.0 ^a ±0.60	9.80 ^a ±0.59
DG 8-10	18.2 ^a ±1.54	14.2 ^c ±1.61	17.5 ^{ab} ±1.62	16.9 ^b ±1.61
<i>Microbiological and immunological traits</i>				
Salmonella count ⁺	10 ^{0.62a} ±10 ^{0.04}	10 ^{0.70a} ±10 ^{0.04}	10 ^{0.64a} ±10 ^{0.04}	10 ^{0.40b} ±10 ^{0.04}
Caecal pH	6.73 ^a ±0.04	6.84 ^a ±0.04	6.79 ^a ±0.04	6.83 ^a ±0.04
Antibody titer* (-log ₂)	9.46 ^b ±0.06	9.54 ^b ±0.06	9.44 ^{ab} ±0.06	9.72 ^a ±0.06

⁺ Salmonella count up to 10^{1.0} was not detected

^{a-c} Means with the same letters within each row of trait are not-significantly different ($P \leq 0.05$)

* Antibody titer expressed as (-log₂)

Means of body weights in INxMA crossbred at 1st, 2nd, 3rd and 4th weeks of age and daily gain during the period from 1-4 weeks of age were significantly higher than reciprocal of MAxIN cross. While, MAxIN cross had higher in daily gains during the intervals 4-8, 8-10 and 1-10 weeks of age than INxMA cross (Table 3). Average of body weight in MA chicks at different ages in this study was generally higher than those corresponding estimates reported by most studies (Abd El-Gawad et al 1979; Abou-El-Ella 1982; Mahmoud et al 1974; Kosba et al 1981; Gohar, 1987 and EL-Sisy 2001).

Means of Salmonella count for genetic groups presented in Table 3 indicate that there were no significant differences between means of the two purebreds of MA and IN at 4th week of age. In spite of, MA strain had slightly higher resistance against Salmonella than IN strain. Comparing between purebreds and crossbreds, it is showed that INxMA cross had the lowest (significant) Salmonella colonization (10^{0.40}) at 4th week of age compared to that obtained in both MAxIN cross (10^{0.64} cfu, not detected) and the two purebreds (Table 3). This may be due to additive and/or non-additive genetic effects of genes. Therefore, it is concluded that Salmonella count in chickens could be decreased by crossbreeding. In an attempt to map the genes responsible for resistance to salmonellosis directly in chickens, Mariani et al (2001) carried out genomic mapping experiments using crossing between line 15I (highly susceptible) and line 6₁ (highly resistant). In these experiments there was a strong association for markers in the distal region of chicken chromosome 5 with resistance, which was confirmed in other crosses between these lines.

Means in Table 3 showed no significant differences between MA and IN purebreds, as well as between the two reciprocal crosses in immunity of chicks against *Salmonella* at 4th week of age. However, birds of INxMA cross had significantly higher immunity of chicks than the two purebreds of MA and IN. This may be due to additive and/or non-additive genetic effects of genes. Linsenmayer et al (1979) showed that the spent culture medium from pooled I₁B6 sub clones (which could be obtained in liter amounts) had an average titer of 6-7 ($-\log_2$) when tested on chicken type I-coated RBCs. On the other hand, it is showed no significant differences between genetic groups for caecal pH in the present study (Table 3).

Crossbreeding effects

Direct additive effect (G^I)

Estimates of G^I and their percentages for the traits studied are given in Table 4.

These estimates were positive for all growth traits and showed highly significant effects only for body weight at 2nd, 3rd and 4th weeks and daily weight gain during interval 1-4 weeks of age. Percentages of these estimates to the means of the two founder parents were ranged from low (2.22%) to high (9.26%) for body weights and from 8.43% to 10.4% for daily gains traits. These results indicate that IN strain gave superiority over MA strain in direct additive of genes for growth traits (Table 4). Thus, it could be used this strain as a sire in crossbreeding programs in Egypt. El-Sisy (2001) found that MA strain favored as sires when crossed with Matrouh and Mandarah strains.

Table 4. Estimates of direct additive effects (G^I) and their percentages for body weight, daily gain and microbiological and immunological traits in crossing of Matrouh and Inshas strains

Trait	$G^I \pm S.E$	$G^I \%^+$	Significance ⁺⁺
<i>Body weight (BW)</i>			
BW1	1.52±0.94	2.22	ns
BW2	4.81±1.6	4.72	**
BW3	7.49±2.5	4.91	**
BW4	11.9±3.4	6.06	**
BW5	14.8±8.6	6.24	ns
BW6	27.9±15	9.26	ns
BW7	29.7±18	8.26	ns
BW8	34.5±22	7.85	ns
BW9	36.7±34	6.49	ns
BW10	57.7±39	8.40	ns
<i>Daily gain (DG)</i>			
DG1-4	0.51±0.14	8.43	**
DG4-8	0.89±0.59	10.1	ns
DG8-10	1.69±1.60	10.4	ns
DG1-10	0.89±0.59	9.1	ns
<i>Microbiological and immunological traits</i>			
Salmonella count	0.08±0.04	12.1	*
Caecal pH	-0.08±0.04	-1.11	ns
Antibody titer	41.1±49	6.32	ns

⁺Percentage computed as $[Estimate\ of\ G^I / (MA+IN)/2] \times 100$

⁺⁺ns= non-significant, *= $P \leq 0.05$ and **= $P \leq 0.01$

Results in Table 4 indicate also that crossing of MA and IN strains was associated with significantly positive effect of G^I on Salmonella count, because it is increased by 12.1% expressing undesirable effect in production of broiler chickens. On the other hand, estimate of G^I for caecal pH was negative and low (-1.11%), but non-significant. For immunity trait, it is showed that insignificant (positive) effect of G^I on antibody titers against *Salmonella typhimurium* at 4th week of age. The immunity of birds was increased by 6.32% (Table 4). Because estimate of G^I was positive, it is concluded that

chicks sired by IN strain would increase the immunity of broilers. There is no available information in literature for effects of G^I on Salmonella count, caecal pH and antibody titers to compare the obtained results in this study.

Maternal breed effect (G^M)

Estimates of G^M and its percentages for the traits studied are given in Table 5. Most of these estimates were negative for growth traits and showing highly significant effects only for body weight at 1st, 2nd, 3rd and 4th weeks of age and daily weight gain during interval 1-4 weeks of age (Table 5). Percentages of these estimates were generally low and ranged from -5.76% to 0.60% for body weights and -4.96% to 1.82% for daily gains traits (Table 5). Negative estimates of G^M obtained in this study indicated that chicks mothered by MA strain are preferred for growth traits compared to chicks mothered by IN strain (Table 3). Thus, it is recommended to use MA dams in crossbreeding programs in Egypt to improve these traits. Also, Iraqi et al (2002) found negative effects of G^M on body weight traits and favored Matrouh dams when crossed Mandarah and Matrouh chickens. They found that maternal effect was highly significant and ranged from -1.40 to -7.73% for daily gain traits.

Table 5. Estimates of maternal effects (G^M) and their percentages for body weight, daily gain and microbiological and immunological traits in crossing of Matrouh and Inshas strains

Trait	$G^M \pm S.E$	$G^M \%^+$	Significance ⁺⁺
<i>Body weight (BW)</i>			
BW1	-1.38±0.66	-2.02	*
BW2	-2.91±1.10	-2.85	**
BW3	-8.79±1.76	-5.76	**
BW4	-7.52±2.37	-3.84	**
BW5	-4.59±6.2	-1.93	ns
BW6	-5.37±11	-1.78	ns
BW7	-0.46±13	-0.13	ns
BW8	-3.43±16	-0.78	ns
BW9	3.42±24	0.60	ns
BW10	-0.36±28	-0.05	ns
<i>Daily gain (DG)</i>			
DG1-4	-0.30±0.10	-4.96	**
DG4-8	0.11±0.42	1.24	ns
DG8-10	0.29± 1.14	1.82	ns
DG1-10	0.01±0.42	0.15	ns
<i>Microbiological and immunological traits</i>			
Salmonella count	-0.12±0.03	-18.2	**
Caecal pH	0.02±0.03	0.29	ns
Antibody titer	-50.0±33	-7.69	ns

⁺Percentage computed as [Estimate of $G^M / (MA+IN)/2] \times 100$

⁺⁺ns= non-significant, *= $P \leq 0.05$ and **= $P \leq 0.01$

Results in Table 5 showed that effect of G^M was highly significant on Salmonella count at 4th week of age. It was decreased the Salmonella count by 18.2%, thus dams of MA strain could be favored in crossbreeding programs in Egypt to decrease the microbial count of Salmonella in broiler chickens. On the other hand, effect of G^M on caecal pH was positive (non-significant) and very low estimate (0.29%). Results given in Table 5 showed that estimate of G^M was negative and moderate (-7.69%) for antibody titers at 4th week of age. It is indicated that dams of IN strain were favored to give high antibody titers compared to MA ones. There is no available information in literature for effects of G^M on Salmonella count, caecal pH and antibody titers to compare the obtained results in this study.

Heterosis effect (H^I)

Estimates of H^I given in Table 6 were mostly positive for growth traits and significant effects only for body weight at 1st, 3rd and 5th weeks of age, but no significant effect for the other traits.

Table 6. Estimates of heterosis effects (H^I) and their percentages for body weight, daily gain and microbiological and immunological traits in crossing of Matrouh and Inshas strains

Trait	$H^I \pm S.E$	$H^I \%^+$	Significance ⁺⁺
<i>Body weight (BW)</i>			
BW1	-2.05±0.94	-2.98	*
BW2	-2.87±1.57	-2.81	ns
BW3	-8.00±2.49	-5.24	**
BW4	-1.17±3.35	-0.87	ns
BW5	18.5±8.6	7.80	*
BW6	23.4±15	7.75	ns
BW7	32.6±18	9.05	ns
BW8	33.4±22	7.70	ns
BW9	39.6±34	6.99	ns
BW10	47.2±39	6.87	ns
<i>Daily gain (DG)</i>			
DG1-4	0.03±0.14	0.50	ns
DG4-8	1.01±0.59	11.3	ns
DG8-10	1.00±1.60	6.15	ns
DG1-10	0.78±0.59	8.03	ns
<i>Microbiological and immunological traits</i>			
Salmonella count	-0.14±0.04	-21.2	**
Caecal pH	0.03±0.04	0.37	ns
Antibody titer	163±49	25.2	**

⁺Percentage computed as $[Estimate\ of\ H^I / (MA + IN) / 2] \times 100$

⁺⁺ns= non-significant, *= $P \leq 0.05$ and **= $P \leq 0.01$

Percentages of these estimates ranged from -5.24% to 9.05% for body weights and 0.5 to 11.3% for daily gain traits. In general, estimates of H^I in this study indicate that crossing of MA and IN strains gave the highest effect of heterosis for growth traits. Khalil et al (1999) and Iraqi et al (2002) found significant heterotic effect on body weight traits in chickens. On the contrary, Hanafi and Iraqi (2001) showed non-significant heterotic effects on daily gain at 8 weeks of age.

For microbiological trait, results presented in Table 6 showed significant effect of H^I on Salmonella count. Estimate of H^I strongly decreased by 21.2%, indicating that crossing of MA and IN strains should be considered to decrease the microbial count of Salmonella in Egyptian broiler chickens. On the other hand, effect of H^I on caecal pH was positive and very low estimate (0.37%). Estimate of H^I found in Table 6 indicates that positive heterotic effect on antibody titers at 4th week of age. It was increased ($P \leq 0.01$) the antibody titer by 25.2%. It is concluded that crossing of MA and IN strains could increase the immunity in broilers against *Salmonella typhimurium*. This is an encouraging factor to improve antibody titers in broilers by crossing. There is no available information in literature for the effect of H^I on Salmonella count, caecal pH and antibody titer traits to compare the obtained results in this study.

Conclusions

- Generally, strain of IN could be used as a sire and MA strain as a dam to improve growth and immunological traits. Thus IN x MA crossbred could be used for increasing growth traits and immunity of chicks against Salmonella (increasing antibody titer), as well as decreasing Salmonella colonization in broilers at the same time.
- Based on maternal breed effect, dams of IN strain could be favored to give the highest antibody titers of broilers against Salmonella typhimurium.
- Based on heterotic effects (significant) on Salmonella count, crossing between MA and IN could be beneficial to: decrease the microbial count of Salmonella, produce healthy meat products from broilers for consumers, and save money used in medication.

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