4. RESULTS AND DISCUSSION

4.1 Effect of dietary protein level on immune response of the experimental chicks:

The effect of feeding different levels of protein on active thyroid immunity was evaluated by the determination of antibody production in response to thyroglobulin antigen stimulation for both female and male Hubbard chicks fed low, medium and high levels of protein.

Antibody formation and titre was assayed in control, adjuvant injected and thyroglobulin immunized groups by three methods:

- Determination of the percentage of T_4 - ^{125}I bound to gamma globulin in serum by ammonium sulphate method.
- The change in relative concentrations of serum gamma globulins by using cellulose acetate electrophoresis.
- The change in serum values of total thyroxine and serum triiodothyronine at various time intervals before and after immunization.

4.1.1. Percentage of T₄-125 I bound to gamma globulin

Data concerning the effect of dietary protein levels on antibody titre of experimental Hubbard chicks are presented in table (2) and illustrated in figures 2 and 3. Obtained results indicated that percentage T_4 - ^{125}I bound to gamma globulin was significantly affected by chick's sex, dietary protein level, thyroglobulin immunization and time intervals (ANOVA Table3).

Regardless of any other factor , female chicks had the higher percentages of T_4 - ^{125}I bound to gamma globulin than males . Grand means mounted (14.77 %) and (12.43) % for female and male chicks, respectively. Similarly , in most experimental groups , females showed higher values of % T_4 - ^{125}I bound to gamma globuin .

Protein level in chick's diet had significant effect on % T_4 - ^{125}I bound to gamma globulin in most experimental groups. Birds fed relatively high level of protein had the highest mean values. While those fed low level of dietary protein had the lowest values. However, chicks recieved medium level of protein were intermediate.

This was quite true in both male and female chicks, percentage T_4 - ^{125}I bound to gamma globulin were 11.62, 14.81, and 15.86% for female chicks fed low, medium and high protein level. While the corresponding values in males were 9.96, 14.06, and 15.58%, respectively.

Thyroglobulin immunization were found to have highly significant effect on antibody formation in most groups (ANONA table 3). Control chicks receiving no treatment had the lowest % T_4 -125 bound to gamma globulin mean values, while chicks immunized with thyroglobulin showed higher immune response than those injected with adjuvant. This was clearly observed in most experimental groups.

This trend has been observed clearly in both male and female chicks. The grand means were 11.72, 11.76, and 17.50% for female of control,

adjuvant injected group and thyroglobulin immunized group, respectively. The corresponding values were 11.79, 12.00, and 16.84% in males, respectively.

Values of antibody titre increased with the progress of time reaching its peak values at the 6^{th} and 8^{th} week after immunization . The grand mean of female chicks before immunization was 9.83 % while it was 11.25 , 14.46 , 15.89 and 16.42 % at the 2^{nd} , 4^{th} , 6^{th} and 8^{th} weeks after immunization. , the corresponding mean values of % T_4 - 125 I bound to gamma globulin in males were 9.28 % before immunization and 10.93 , 13.87 , 15.91 and 16.50 % thereafter for the same intervals, respectively.

Significant interactions effects were observed between protein level x time intervals and immunizations x time intervals (ANOVA table 3) which indicated that protein levels in diet and immunization treatment values were affected by various time intervals before and after immunization.

It becomes well known that avian species particulary chickens are considered an important models for studying viral oncogensis including immune responses against both causal agents and tumors (Schat . 1991).

Knowledge about avian immune system is relatively limited, most important researches directed towards understanding of its biology has been conducted in mammalian species (Qureshi and Miller, 1991).

With the increasing need for the development of vaccines against many poultry diseases it is important to understand host mechanims in protection against diseases (Lillehaj, 1991).

Antibodies have been demonstrated to prevent initial invasion of microbes (Isobe et al., 1989), However, the recognition of type and level of antibody in serum became a very important tool in diagnosis of many diseases especially autoimmune disease that produced by the body itself.

As already known that the immune response of chicken's is influenced by a number of nutrients, mainly protein. The present study tried to give lights on the antibody formation and to how extent did the protein level in diet affect it's production and level in both female and male Hubberd chicks.

As a function of treatment, the present study revealed that all chicks produced antibodies about the $2^{\rm nd}$ week after immunization. At this interval no clear difference in the percentage of T_4 - ^{125}I bound to gamma globulin was observed within the experimental levels of protein studied in this research.

With the progress of time after immunization, remarkably increase in antibody titre was noted at the 4^{th} , 6^{th} and 8^{th} interval in thyroglobulin immunized group than the control or adjuvant injected group. This increase may be due to the primary and secondary immuization by thyroglobulin as foreign antigen.

Significant results obtained due to sex, showed that females had the higher percentage of T_4 - ^{125}I bound to gamma globulins than males at all the time intervals particularly at the peak time of 4th and 6th week after

immunization . This may lead to the observation that females chicks produce higher levels of antibodies than males in response to invading antigens . This result was in agreement with (Glick et al., 1983) who reported that the proportional weight of Bursa of Fabricious (responsible for secreting B lymphocytes) in chicks to body weight was higher in females than in males . It averaged 0.39 % in females and 0.36 in males at the 6th weeks of age. However , the ability of females Fabricious gland to produce immunoglobulin is considered to be more than males .

IgM levels in serum are slightly higher in women than in age matched men, apparently due to X- linked determinants of base line levels (Peter and Gorevic, 1983).

Antibody production was significantly affected by the level of protein in diet. Thus dysfunction in any sequence of the host protective machaniams may be affected with protein deficiences or excesses in diet and in turn affect the immunologic responsivness (Asheffy and Williams , 1979).

The present finding revealed that low level of protein in diet has the lower antibody titre in most of the experimental groups . while higher levels of dietary protein leads to the production of more antibodies . This may be attributed to that low protein intake by chicks not only delay the overall growth of the bird but also delay the normal atrophy of the lymphoid organs responsible of immunoglobulin production . As already reviewed that total lymphocyte counts are depressed in protein restricted birds , most proportion and absolute number of T cells is decreased (they do function in conjunction

with antibody response). However, this also may explain that higher levels of protein in diet increase the availability of amino acids in serum needed to activate the immune system. Asheffy and Williams, (1979) concluded that protein deprivation affects specific amino acids in diet which appears to affect primarily humoral response in chickens.

In addition, protein as a nutrient is considered to be of a vital importance to cellular nucleic acid synthesis and cell proliferation. Deficiency of protein directly involveed in nucleic acid metabolism, cell division and or protein synthesis, would have deleterious effects on all forms of immunologic activity.

These results are in agreement with (Glick et al., 1981; Oyeijde et al., 1985; Payne et al., 1988 and Bryan and Jensen, 1989). Payne et al., (1990) concluded that protein as well as type of vaccination had significant effects on antibody titre, however, protein restricted group of birds were lower in antibody formation especially $\rm I_g \ G$.

The present study are in agreement with those obtained by Cooper et al., (1974) and Bell et al., (1976) that modertly severe protein deficiency may cause suppression in humoral immune response. Concerning the effect of low protein level in diet on immune response, Glick et al., (1983) concluded that lower dietary protein causes differences in total white blood cells, absolute lymphocytes or absolute heterophils during the first weeks of age. Significant lower number of lymphocyte was observed in protein restricted groups in the study of Payne et al., (1990) who concluded that protein deficiency in chicken's diet lowers their overall immune status.

The present results are in partial disagreement with those reported by Oyeijd et al., (1984) and Nayak and Rai, (1986) indicating that antibody production in serum increased with lower level of protein in diet. This may be due to the difference in challange faced by birds or to the variations of antigens and level of protein used by chicks.

Another group of investigators, on the other hand, reported no apparent change in gamma globulin production as a function of low or high levels of protein in diet (Hoffenberg et al., 1966 and Mohamed et al., 1980).

The maximum thyroglobulin antibody titre was reported to vary between the 4^{th} to 8^{th} weeks after primary immunization (Schumacher and Premachandra , 1968 , Pogoriler <u>et al.</u>, 1971 and Ibrahim and Premachandra , 1972). Maximum antibody titre observed in the present study was at 4^{th} , 6^{th} and 8^{th} week after immunization .Odette , (1986) and Ibrahim <u>et al.</u>, (1988) showed another peak attained about the 12^{th} week after thyroglobulin immunization of male chicks . This may be attributed to the route and frequency of immunization, emulsification of the immunizing antigen and kind of recepient .

The most common findings is the increase in gamma globulin fraction as sequence of immunity either, active ar autoimmune.

In conclusion, although antibodies have been found to be associated with gamma globulin, it is quite evident that the total quantity of gammaglobulin is affected by the level of protein in diet. However, the immuno comeptence have been shown to be a sensitive indicator of adequancy of protein restriction

and should serve as a beneficial addition to the more common use of metabolic profiles in their evaluation.

As the absolute number of (B)lymphocytes are affected greatly by the level of dietary protein, it might be the case that using moderat littel higher protein levels in diet by breeders is important to activate the sequences of immune system and to help the birds to persist for longer time.

Concerning the use of chicken's as animals for antibody production, this raise the question about the importance of considering various intervals of antibodies peak time of the specified antigen.

Table (2) Effect of dietary protein levels on the immune response ${\%T_4 - 125}_1$ bound, to gamma globulin ($\overline{X} \pm S.E.$) of Female and Male treated chicks at various time intervals before and after thyroglobulin immunization.

% T ₄ -125, BOUND TO GAMMA GLOBULIN									
Protein				fter immu					
level	Group	Before	2 nd	4 th	6 th	8 th			
FEMALE									
	Cont	7.69 <u>+</u> .46	9.34 <u>+</u> .62	11.13±.40	11.32 <u>+</u> .27	12.53+.80			
Low	Adj	7.58 <u>+</u> .42	9.26±.76	11.29 <u>+</u> .36	11.45 <u>+</u> .35	12.23+.35			
	Tg	8.51±.33	11.50±.62	16.98 <u>+</u> .42	16.78 <u>±</u> .21	15.34+.85			
	Cont	9.79 <u>+</u> .34	10.59 <u>+</u> .59	13.23 <u>+</u> .23	12.82 <u>+</u> 1.02	13.50+.17			
Medium	Adj	8.78 <u>+</u> .32	10.92 <u>+</u> .73	12.38±.73	13.19 <u>+</u> .60	13.53+.68			
	Tg	9.22 <u>+</u> .39	13.93 <u>+</u> .28	21.73±.59	24.90 <u>+</u> 106	24.68+1.73			
		ł		:	14.72 <u>+</u> .65				
High	_					14.46+1.09			
	Tg !	10.04 <u>±</u> .93	12.28±.70	MALE	27.10±1.14	26.29+1.62			
		7.60 04	0.40 . 72		10.09 . 24	12.39+.56			
	Cont	7.62 <u>+</u> .04	8.48 <u>+</u> .73	 I	10.08±.34				
Low	Adj	8.14 <u>+</u> .55							
	Tg	7.87 <u>+</u> .43	9.56 <u>+</u> .48	15.58 <u>+</u> .34	14.47 <u>+</u> .54	14.97+.45			
	Cont	9.12 <u>+</u> .84	11.47 <u>+</u> .46	13.04 <u>+</u> .13	13.48 <u>+</u> .58	14.09+.22			
Medium	Adj	8.63 <u>+</u> .16	11.04 <u>+</u> .32	12.09 <u>+</u> .47	13.65±.34	14.84+.68			
	Tg	9.16 <u>+</u> .72	12.09 <u>+</u> .96	20.20 <u>+</u> .76	23.10 <u>+</u> 1.29	24.92+1.68			
	Cont	10.77 <u>+</u> .75	12.21 <u>+</u> .43	13.88 <u>+</u> .98	14.82 <u>+</u> 1.09	14.79+1.36			
High	Adj	11.16 <u>+</u> .45	12.45 <u>±</u> .21	13.13 <u>±</u> .37	15.28±.16	15.09+1.25			
	Tg	19.09 <u>+</u> .44	14.09 <u>+</u> .68	24.42 <u>+</u> .08	26.98±1.20	25.56+1.39			

N.B Cont = control group, Adj = adjuvant injected group. Tg = Thyroglobulin immunized group

Table (3) Analysis of variance for Data represented in table (2)

source of variation	D.F	sum of	Mean	F
Sex (S)	1	68.911	68.911	17.723***
Protein level (P)	2	160.477	80.238	20.636***
Immunization (M)	2	413.988	206.994	53.236***
Interval (I)	4	1721.979	430.494	110.718***
SxP	2	6.606	3.303	.850
SxM	2	10.088	5.044	1.297
SxI	4	27.821	6.955	1.789
P x M	4	18.406	4.601	1.18
PxI	8	146.639	18.329	4.714**
MxI	8	359.991	44.998	11.573***
Remainder	637	2476.799	3.888	

^{*} P < 0.05

^{**} P < 0.01

^{***} P < 0.001

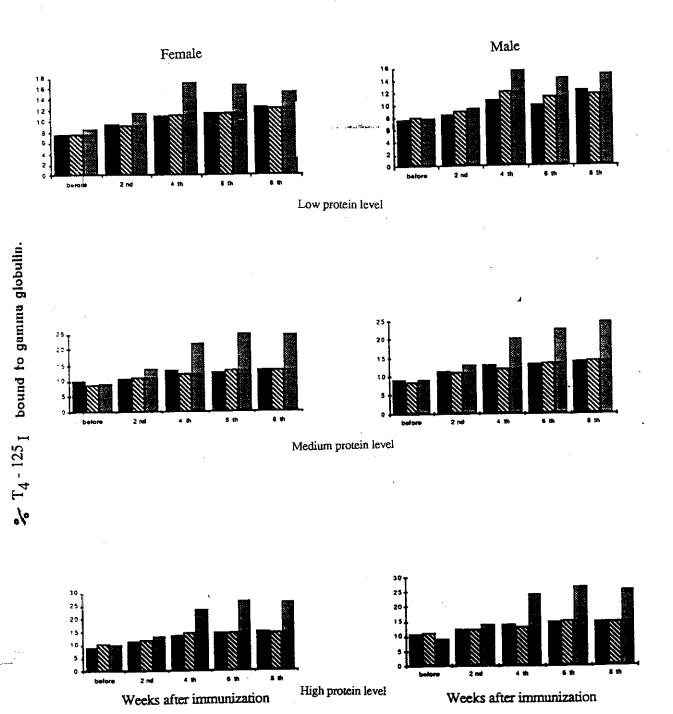
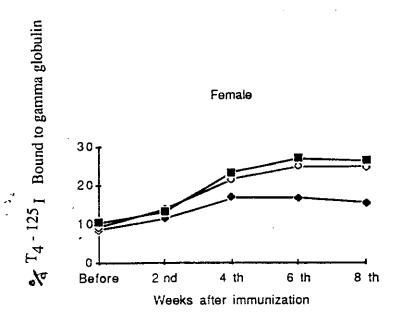


Fig (2) Percentage of T_d 125 I ound to gamma globulin (means) of Fernale and Male chicks at low, medium and high levels dietary protein before and after immunization.

	Cont
Z	Adj
	Тg



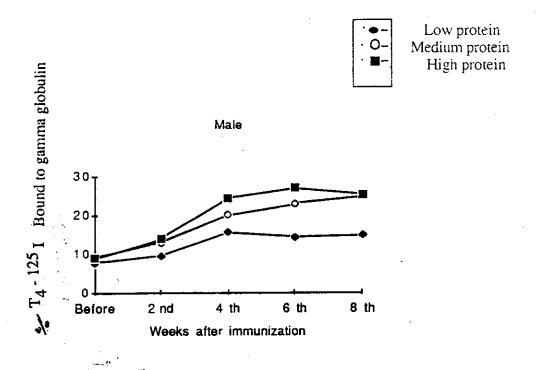


Fig (3) Percentage of T_4 - 125 I bound to gamma globulin (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

4.1.2 Electrophoretic fractionation of serum proteins

Qualitative analysis of serum proteins was carried out using cellulose acetate electrophoresis to determine the relative concentration of each fraction particularly gamma globulin.

4.1.2.1 Serum albumin (Alb.)

Data concerning the effect of dietary protein level on for both female and male chicks at various time intervals before and after immunization for low, medium and high levels of dietary protein are prsented in tables (4, 5 and 6), respectively and illustrated in figures 4 and 5.

The data obtained showed significant effect due to sex, dietary protein level and time intervals on serum albumin content (ANOVA table 7).

Over all means of serum albumin in female and male chicks had almost similar values being 1.70 g / dl and 1.66 g/dl for females and males, respectively.

Protein level in chicks diet had significant effect on albumin concentration in serum. With the increase in protein level in diet, there was always on average, an increase in serum albumin. Serum albumin in females was 1.59, 1.68 and 1.98 g/dl for chicks fed low, medium and high levels of dietary protein, while the corresponding values in males were 1.56,1.69 and 1.72 g/dl, respectively.

Increased mean values of serum albumin were noted as effect of age. This was true in all experimental groups of birds. Female values were increased consistently from 1.54 g/dl before immunization to 1.63, 1.71, 1.75 and 1.77 g/dl at the $2^{\rm nd}$, $4^{\rm th}$, $6^{\rm th}$, $8^{\rm th}$ week after immunization, while, the corresponding values in males were 1.60 g/dl before immunization and 1.60,1.66, 1.70 and 1.77 g/dl, respectively for the same intervals.

Significant interaction effect was noted between sex and immunization (ANOVA table7) indicated that immunization affect female chicks in different way than males.

Serum Globulins:

4.1.2.2 Alpha - globulin (α - glob)

Data presented in table (4,5 and 6) represent variations of serum alpha - globulin g/dl mean values as affected by dietary protein level and immunization for both female and male chicks at various time intervals and illustrated in figures 6 and 7.

Significant effects due to sex, protein level and time intervals were observed to affect serum mean values of alpha - globulin g/dl (ANOVA table?)

Female chicks had higher mean values (4.9 g/dl) than males (4.1 g / dl). In spite of the significante effect of dietary protein level on serum alpha globulin g/dl, little differance was noted between the three main levels of protein. Female chicks attained 0.65, 0.50 and 0.52 g/dl for birds fed low, medium and high level of dietary protein. While the corresponding values in males were 0.44, 0.48 and 0.46 g/dl, respectively.

As effect of age, alpha globulins mean values was slightly increased from before to the 8th week after immunization, A significant interaction effect between protein level and time intervals was noted (ANOVA table 7).

4-1-2-3 Beta globulin $(\beta$ -glob)

Data concerning Beta globulin (g/dl) are presented in tables (4, 5 and 6) for chicks fed low, medium, and high levels of protein in diet, and illustrated in figures 8 and 9.

No significant differences were obtained in beta globulin (g/dl) mean values due to the effect of any of the tested factors (ANOVA table 7), However, no consistent pattern was noted in serum Beta globulin (gldl) as effect of feeding chicks different levels of protien in diet.

Female grand means were 0.98, 0.81 and 0.83 g/dl for chicks fed low, medium and high levels of protein. While the corresponding values in males were 0.84, 0.84, 0.80 g/dl, respectively.

Slight flactuations in Beta globulin mean values were noted between the control, adjuvant injected groups or thyroglobulin immunized groups. The same trend was observed with the progress of time at any time interval .

4-1-2-4 Gamma- globulin (γ - glob).

The increase in relative concentration of serum gamma- glabulin g/dL is considerd to be another reflection for antibody formation. No significant effect due to immunization had been noted for both Alpha or Beta globulin within the time of antibody formation .

Data Concerning the variations of gamma globulin mean values are presented in table (4, 5 and 6) for low, medium and high levels of protein in diet for both female and male chicks at various time intervals before and after immunization, and illustrated in figures 10 and 11.

Significant differences due to the main effects of protein level in diet, immunization and time intervals were observed (ANOVA table 7).

Protein levels in diet had remarkable effect on gamma globulin concentration measured by cellulose acetate electrophoresis in most experimental groups. Chicks fed higher levels of protein in diet attained the higher levels of serum gamma globulin and those fed low levels of dietary

protein attained the lowest values. Chickes recieved medium levels of protein were intermediate. This was quite true in both female and male chicks.

Gamma globulin mean values were 1.29, 1.37 and 1.48 g/dl for female chicks fed low, medium and high level of protein in diet, while the corresponding values were 1.21, 1.36 and 1.43 g/dl in males, respectively.

Significant difference due to immunization were found to affect the concentration of gamma globulin mean values in serum. Control chicks reciveing no immunization treatment had the lowest mean values of gamma globulin, while thyroglobulin immunized group attained the highest values than those injected with adjuvant. This was also observed in most experimental groups of both female and male chicks. The grand mean values of gamma globulin were 1.26, 1.31 and 1.46 g/dl for female chicks of control, adjuvant and thyroglobulin immunized groups, respectively while the corresponding values in males were 1.22, 1.32 and 1.41 g/dL, respectively.

Values of gamma globulin increased with the progess of time attaining its peak values at the 4^{th} , 6^{th} week after immunization, Females mean values of gamma globulin were 1.06 gldl before immunization and 1.30, 1.50, 1.43 and 1.43 g/dl at the 2^{nd} , 4^{th} and 6^{th} week after immunization, while it was 1.03 g/dl, in male before immunization and 1.31, 1.43, 1.51and 1.39 g/dl, respectively after immunization.

Analysis of variance revealed siginficant effect due to the interaction between protein level and time intervals (ANOVA table 7) indicating that dietary protein level affect gamma globulins concentrations in serum differently at each interval.

One of the main factors affecting the level of each protein fraction present in fowl serum or plasma rather than breed, age, environment and physiological state is the exposure to antigens. The amount of each fraction reflects a balance of synthesis and catabolism of the contributing serum protein.

The present results revealed that the chicken's immune sera are separated to albumin, alpha, beta beside gamma globulin, It has been shown that there was an increase in relative concentration of albumin and gamma globulin with the progess of time, which may be attributed to the fact that most serum antibodies have been found to be associated with gamma globulin fraction and to lesser extent to alpha or beta globulin, which has not been affected by immunization. The increase in albumin values with age may be attributed to the increase in serum total protein, while the increase in gamma globulin concentration in serum with the progress of time may be attributed to the increase in naturally occuring gamma globulin.

Higher levels of gamma globulin in the present study were mostly associated with feeding the chickes higher levels of protein in diet. However lower concentration of gamma globulin was also associated with lower dietary protein level. Similar results obtained by using ammonium sulphate method for measuring the percentage of T_4 - 125 I bound to gamma globulin.

The increase in serum gamma globulin levels as effect of immunization was in agreement with the results of Micheal and David, (1983) who reported that the increase in gamma globulin region reflects hyperimmunized state or chronic infection. Hyperglobulinemia is an indicater of dysproteinemia and autoimmune disorders.

Immunoelectrophoresis affords a more positive means of identifying the various proteins, providing that the appropriate specific antigens are avilable in serum and induce precipitation and antibody formation (Butler, 1971).

Twelve proteins have been identified in fowl serum by this techniques (Tueen et al., 1966) and the presence of gamma globulin has been demonstrated.

It may be concluded that antibodies are not always strictly confined to gamma globulin. They may be associated also with other globulins in $gamma_{(1)}$ - fraction or between $beta_{(2)}$ and gamma - globulin regions, and different types of antibodies may have different physiochemical properties (Moore, 1960)

Alpha globulin represent the acute phase reactants. Stricking elevation in this region of the electrophoresis band may be seen in acute and chronic inflammatory states. While elevation in beta-globulin may be indicative of iron deficiency state or hyperlipidemia, decreased levels are seen in nutritional disorders. (Micheal and David, 1983).

Flactuations in either alpha or beta region which had been observed in the present study may be indicative to the insignificant effect of imimunization in our case.

Concerning the adjuvant injected groups of chicks which mostly had little higher values of gamma globulin than control groups, it could be stated that adjuvant has the capacity to stimulate the cells of the antibody system in non specific manner. However, adjuants are able to handle antigens effectively.

In agreement with the present findings are those data presented by Adams et al., (1991) indicating no significant interaction effect between diet and sex on serum gamma globulin level.

In his report Oyeijde, (1984) concluded similar results on feeding different levels of crude protein in diet to chicks immunized against Newcastle diseases, higher antibody titre was associated with higher protein level in diet (22-24%), while lower titre was observed with (17-18%) protein in diet. However, the same author, (1985), partially disagreed with the present study, he

reported that chicks fed 12% protein level in diet had significantly higher antibody titre after lasota. This contradiction may be attributed to the difference in antigens used.

In Conclusion, It is now recognized that the role of nutritional factors in aquired immune response became evident. The reduction in dietary protein level may affect the absolute mumber of B-lymphocyte which are considerably reduced and is reflected in a faliour of lymph nodes to increase in cellularity on antigenic challenge, this in turn affect the antibody production. Deficiency of essential and non essential amino acids may affect the formation of antibodies itself (mainly sulpher containing amino acids). It may also decrease phagocytic activity and leucopenia.

However, It may be of a great practical use to pay attention to the protein level in diet and to apply active immunization against the common poultry viral and bacterial diseases, to increase the level of circulating T and B cells, responsible for antibody formation and memory cells specified to different kinds of antigens, since immune system like nervous system can remmber.

Table (4) Effect of Low dietarty protein levels on different serum protein fractions: (Albumin, Alpha, Beta and gamma globulin g/dl) x +S.E using cellulose acetate electrophoresis of female and male chicks at various time intervals before and after thyroglobulin immunization.

Protein fractions (Alb ,α,β and γ globulins g/dL) Weeks after immunization							
Group	Protein	Before	2 nd	4 th	6 th	8 th	
				ALE			
	Alb	1.32 ±.02	1.65 ±.02	1.58 ±.03	1.72 <u>+</u> .06	1.73 ±.02	
Cont	α - glob.	$0.30 \pm .07$	0.50 <u>+</u> .01	0.59 <u>+</u> .04	0.46 ± .01	$0.54 \pm .01$	
Cont	β - glob.	0.40 <u>+</u> .02	0.88 ± .05	0.90 ± .06	$0.98 \pm .01$	$0.96 \pm .05$	
	γ - glob .	0.83 <u>+</u> .02	1.06 <u>+</u> .04	1.28 ± .09	1.14 ± .04	1.23 ± .05	
	Alb.	1.31 <u>+</u> .11	1.54 <u>+</u> .06	1.59 ± .04	1.71 ± .11	1.72 ± .02	
A 41:	α - glob.	0.34 <u>+</u> .03	0.35 ± .01	$0.55 \pm .04$	0.57 ± .05	$0.68 \pm .01$	
Adj	β - glob.	0.43 <u>+</u> .02	0.69 ± .09	$0.89 \pm .01$	0.94 ± .02	0.98 ± .03	
*	γ - glob.	0.95 <u>+</u> .07	1.09 ± .02	1.31 ± .02	1.30 ± .01	1.39 ± .04	
	Alb	1.53 <u>+</u> .02	1.49 ± .09	1.59 ± .07	1.70 ± .03		
	α - glob.	0.35 <u>+</u> .02	$0.41 \pm .02$	0.44 ± .02	$0.47 \pm .02$		
Tg	β – glob.	0.54 <u>+</u> .03	$0.87 \pm .05$	$0.65 \pm .03$	$0.65 \pm .11$	$0.80 \pm .09$	
	γ - glob.	0.91 <u>+</u> .05	$1.30 \pm .13$	1.68 ± .06	1.56 ± .08	1.51 ± .09	
	1 6100.	<u> </u>	MALE				
	Alb	1.31 <u>+</u> .01	1.45 ± .04	1.41 <u>+</u> .04	1.58 ± .07	1	
Cont	α - glob.	0.32 <u>+</u> .10	$0.41 \pm .02$	$0.36 \pm .01$	L	1	
Com	β – glob.	0.41±.02	$0.60 \pm .01$	$0.57 \pm .05$	1	l	
	γ - glob.		1.19 ± .01	0.99 ± .01	1.35 ± .02	1.13 ± 0.5	
	Alb.	1.48±.01	1.58 ± .02	1.55 ± .05	1.68 ± .11		
A 4:	α - glob.	1		1	0.42 ± .09		
Adj	β - glob.	_ <u>_</u>	1	$0.69 \pm .09$	$0.80 \pm .09$	1	
	γ - glob	1	1	1	1.35 ± .23	2 1.17 ± .09	
	Alb	1.63 ± .05	1.53 ± .04	1.51 ± .0	3 1.65 ± .1		
T-	α – glob	1	1 04		1 0.47 ± .0		
Tg	β - glob		· I _	1 _		1	
	γ - glob		1		1 1.54 ± .1	$3 1.47 \pm .1$	

Table (5) Effect of Medium dietarty protein levels on different serum protein fractions; (Albumin, Alpha, Beta and gamma globulin g/dl) $\vec{x} \pm S.E$ using cellulose acetate electrophoresis of female and male chicks at various time intervals before and after thyroglobulin immunization.

		<u> </u>					
Protein fractions (Alb , α , β and γ globulins g/dL)							
C	Duntain	Before	Weeks after immunization				
Group	Protein	DCIOIC	2 nd	4 th	6 th	8 th	
			FEMALE	•			
<u></u>	Alb	1.66 ± .01	1.50 ± .10	1.73 ± .05	1.79 <u>+</u> .03	1.73 <u>+</u> .08	
Cont	α - glob.	0.41 ± .02	0.38 <u>+</u> .04	0.61 <u>+</u> .04	0.55 <u>+</u> .06	.55 <u>+</u> .08	
į	β - glob .	$0.80 \pm .05$	0.72 ± .08	$0.84 \pm .04$	0.86 ± .06	0.97 ± .11	
	γ - glob .	1.09 ± .10	1.13 <u>+</u> .11	1.30 <u>+</u> .06	1.43 <u>+</u> .06	1.43 ± .07	
	Alb.	1.71 <u>+</u> .04	1.64 ± .13	1.67 ± .03	1.75 <u>+</u> .04	1.73 ± .07	
Adj .	α - glob .	0.45 ± .01	0.35 <u>+</u> .01	0.54 ± .08	0.47 ± .01	$0.60 \pm .03$	
	β - glob .	0.80 <u>+</u> .06	0.56 ± .02	0.71 <u>+</u> .10	0.97 ± .01	0.98 ± .12	
	γ - glob .	1.16 <u>+</u> .04	1.25 ± .04	1.48 ± .10	1.46 ± .01	$1.47 \pm .03$.	
	Alb	1.42 ± .03	1.73 <u>+</u> .08	1.70 ± .06	1.64 ± .08	1.85 ± .08	
Tg	α - glob.	0.62 <u>+</u> .04	0.53 ± .06	0.52 ± .05	0.48 ± .01	0.49 ± .03	
_	β – glob .	$0.60 \pm .02$	0.74 <u>+</u> .08	0.78 ± .12	0.89 <u>+</u> .07	0.91 ± .11	
	γ-glob.	0.95 ± .04	1.45 ± .08	1.62 <u>+</u> .10	1.83 <u>+</u> .07	1.55 <u>+</u> .07	
			MALE	_			
	Alb	1.72 ± .01	1.50 <u>+</u> .04	1.69 <u>+</u> .04	1.68 ± .05	1.86 <u>+</u> .09	
Cont	α - glob.	$0.37 \pm .02$	0.59 <u>+</u> .04	0.42 <u>+</u> .05	0.39 <u>+</u> .02	0.42 <u>+</u> .02	
	β – glob .	0.65 <u>+</u> .05	0.81 <u>+</u> .01	0.82 <u>+</u> .10	0.71 <u>+</u> .03	0.64 <u>+</u> .02	
	γ-glob.	0.89 <u>+</u> .01	1.15 ± .08	1.24 ± .04	1.35 ± .03	1.29 ± .06	
	Alb.	1.53 <u>+</u> .16	1.76 ± .08	1.63 ± .05	1.75 ± .05	1.79 ± .05	
Adj	α - glob .	0.41 ± .01	$0.56 \pm .02$	0.60 ± .04	0.60 ± .01	0.57 ± .06	
	β - glob .	0.47 ± .02	0.85 <u>+</u> .05	0.82 ± .05	0.96 ± .01	0.89 ± .13	
,	γ - glob.	0.99 ± .03	1.39 ± .05	1.48 ± .03	1.50 <u>+</u> .02	1.47 ± .02	
	Alb	1.60 ± .01	1.68 ± .07	1.64 ± .02	1.72 ± .09	1.80 <u>+</u> .04	
Tg	α – glob.	0.48 <u>+</u> .05	$0.44 \pm .03$	$0.35 \pm .06$	$0.47 \pm .01$	0.56 <u>+</u> .04	
-8	β - glob .	0.67 ± .13	0.63 <u>+</u> .02	0.48 ± .09	0.78 ± .07	0.66 <u>+</u> .04	
	γ - glob .	1.32 ± 013	1.43 ± .11	1.68 ± .15	$1.80 \pm .07$	1.47 ± .04	

Table (6) Effect of High dietarty protein levels on different serum protein fractions: (Albumin, Alpha, Beta and gamma globulin g/dL) $\bar{x}\pm S.E$ using cellulose acetate electrophoresis of female and male chicks at various time intervals before and after thyroglbulin immunization.

	Protein fractions (Alb , α , β and γ globulins g/dL)							
_		D. C	W	eeks after imm	unization			
Group	Protein	Before	2 nd	4 th	6 th	8 th		
FEMALE								
	Alb	1.66 ± .10	1.77 ± .03	1.93 ± .08	1.89 ± .13	1.90 <u>+</u> .04		
Cont	α - glob.	$0.42 \pm .03$	$0.47 \pm .02$	0.56 ± .08	0.59 <u>+</u> .05	0.47 ± .02		
	β - glob .	0.84 ± .02	0.65 ± .04	0.88 ± .07	0.85 <u>+</u> .07	$0.87 \pm .03$		
	γ - glob .	$1.23 \pm .10$	1.31 ± .02	1.57 <u>+</u> .06	1.49 ± .04	1.42 ± .07		
	Alb.	1.65 <u>+</u> .07	1.71 <u>+</u> .14	1.78 <u>+</u> .04	1.86 ± .02	1.79 <u>+</u> .12		
Adj ,	α - glob .	0.40 <u>+</u> .02	0.76 ± .02	0.62 <u>+</u> .05	$0.65 \pm .02$	0.65 ± .02		
,,	β - glob .	0.97 <u>+</u> .08	0.95 ± .04	0.89 <u>+</u> .09	0.89 ± .05	0.95 <u>+</u> .03		
¥	γ - glob.	1.31 <u>+</u> .04	1.50 ± .01	1.53 ± .12	1.51 ± .10	1.43 ± .07.		
	Alb	1.62 ± .10	1.69 ± .10	1.89 ± .08	1.81 ± .10	1.80 ± .08		
Tg	α - glob.	0.40 ± .02	0.52 ± .02	0.52 ± .04	0.56 ± .07	0.48 ± .03		
.5	β – glob.	0.73 <u>+</u> .04	0.65 ± .05	0.81 ± .08	0.72 ± .12	0.81 <u>+</u> .09		
	γ - glob .	1.16 ± .13	1.61 ± .10	1.84 ± .09	1.71 <u>+</u> .13	1.47 <u>+</u> .08		
	<u>, , , , , , , , , , , , , , , , , , , </u>	· ·	MALE					
	Alb	1.62 ± .02	1.61 ± .04	1.89 <u>+</u> .04	$1.77 \pm .05$	1.81 ± .03		
Cont	α - glob.	$0.36 \pm .01$	0.51 ± .05	0.58 <u>+</u> .06	$0.56 \pm .02$	$0.37 \pm .02$		
	β – glob .	0.67 ± .10	0.85 ± .04	0.97 <u>+</u> .05	0.87 <u>+</u> .08	$0.62 \pm .02$		
	γ - glob.	0.88 <u>+</u> .01	1.38 ± .05	1.56 ± .01	1.48 ± .02	1.34 ± .03		
	Alb.	1.79 <u>+</u> .09	1.66 ± .09	1.75 ± .04	1.56 <u>+</u> .05	1.73 ± .05		
Adj	α - glob.	0.47 <u>+</u> .02	0.50 ± .02	0.59 ± .03	0.45 <u>+</u> .02	$0.41 \pm .03$		
1.3.	β - glob.	0.73 ± .08	0.64 ± .05	0.91 ± .01	0.95 <u>+</u> .01	0.76 ± .06		
	γ - glob.	1.08 ± .19	1.66 ± .01	1.67 ± .02	1.78 ± .04	1.58 ± .19		
	Alb	1.72 ± .02	1.71 ± .08	1.88 ± .05	1.95 ± .02	1.88 ± .11		
Тд	α – glob.	$0.41 \pm .03$	$0.53 \pm .03$	$0.57 \pm .01$	0.52 <u>+</u> .02	$0.53 \pm .07$		
^6	β - glob.	0.81 ± .08	$0.85 \pm .02$	0.84 ± .02	0.76 <u>+</u> .05	$0.85 \pm .08$		
	γ - glob.	1.08 ± .19	1.66 ± .01	$1.67 \pm .02$	1.78 <u>+</u> .04	1.58 ± .09		

Table (7) Analysis of variance for Data represented in table (4,5,6).

Source of variation	D.F	Albumin mean squares	α - glob mean squares	β- glob mean squares	γ - glob mean squ ares
Sex (S)	1	.168*	.057**	1.136	.063
Protein level (p)	2	.290***	.065***	.990	.468***
Immunization (M)	2	.005	.016	.725	.303***
Interval (I)	4	.123**	.082***	.163	1.522***
SxP	2	.037	400 1	.153	.060
SxM	2	.117*	.005	.518	.075
SxI	4	.051	.005	.262	.025
PxM	4	.010	.004	.449	.008
PxI	8	.043	.023**	.542	.052
MxI	8	.015	.009	.350	.021
Remainder	172	.031	.008	.356	.026

^{*} P < 0.05

^{**} P < 0.01

^{***} P < 0.001

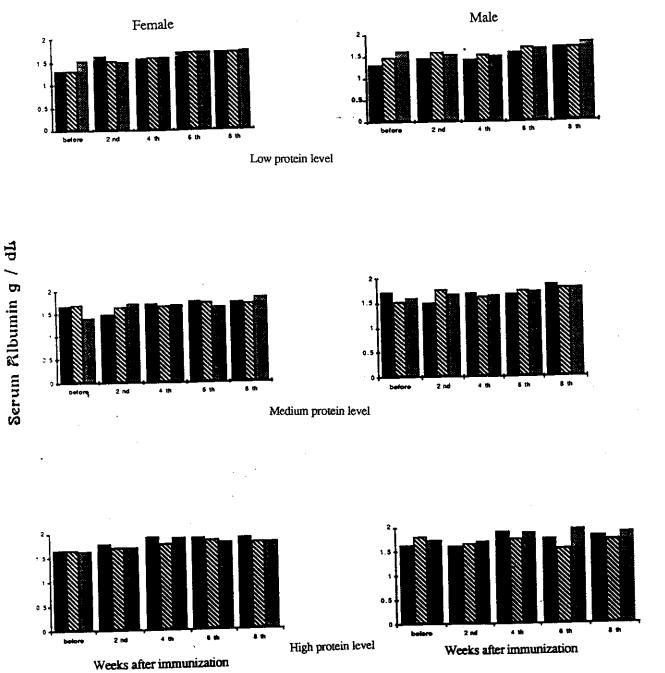


Fig (4) Serum albumin (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.



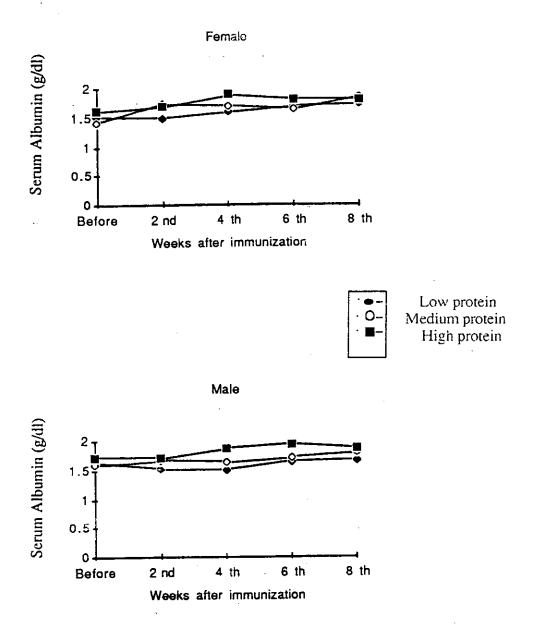
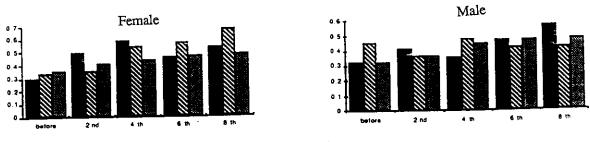
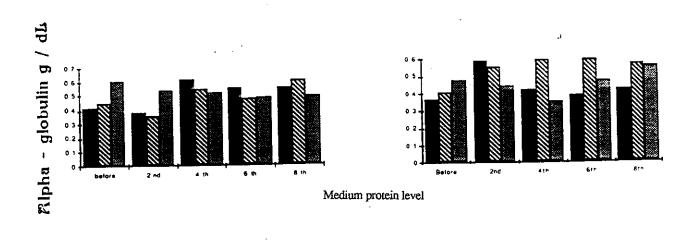


Fig (5) Serum Albumin (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals



Low protein level



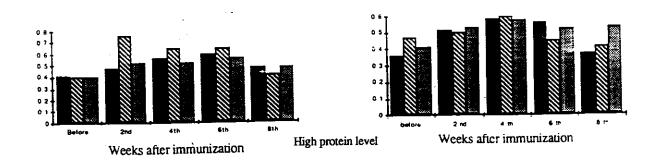
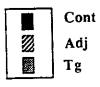


Fig (6) Serum Alpha - globulin (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.



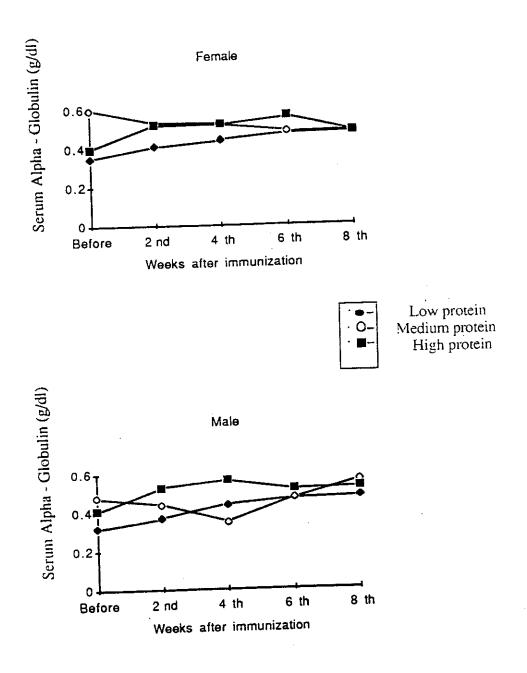
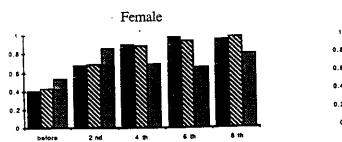
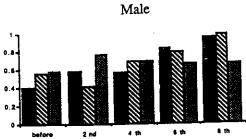
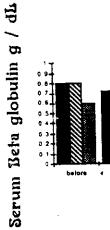


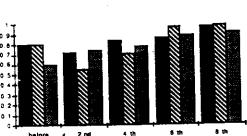
Fig (7) Serum Alpha-globulin (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

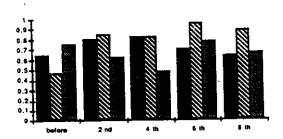




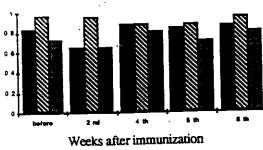
Low protein level

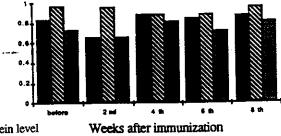






Medium protein level





weeks after minimumzation

High protein level

Fig (8) Serum Beta - globulin (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.

Cont

Adj

Tg

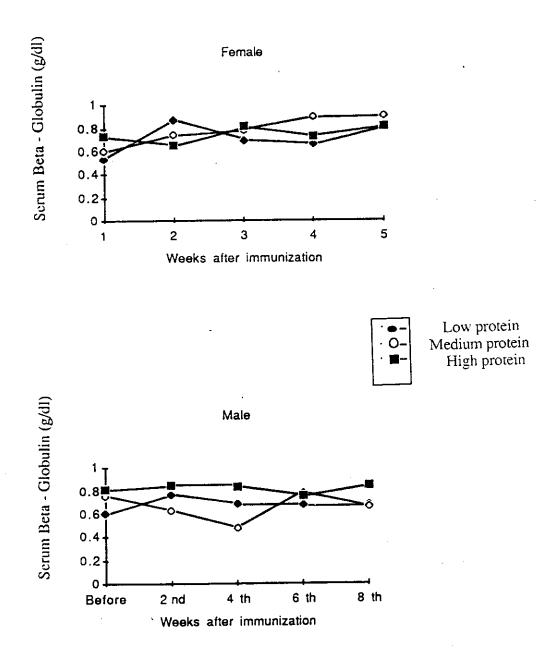


Fig (9) Serum Beta-globulin (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

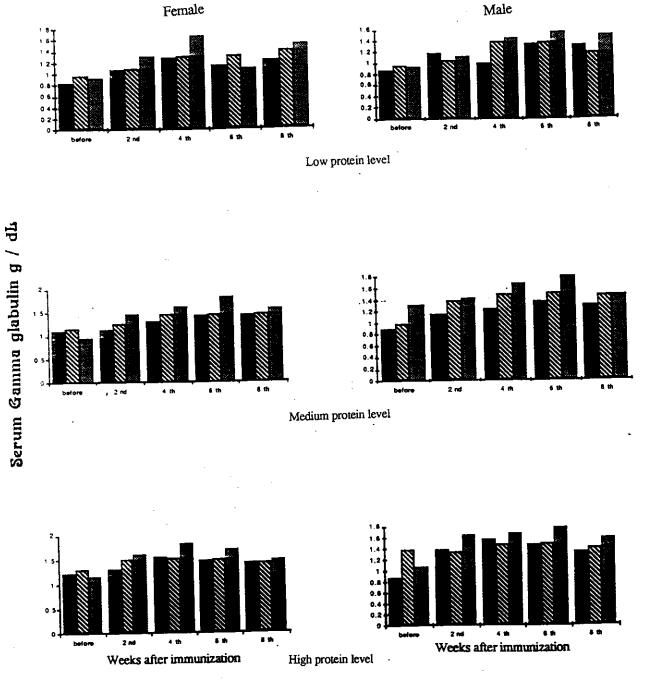


Fig (10) Serum gamma globulins (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.



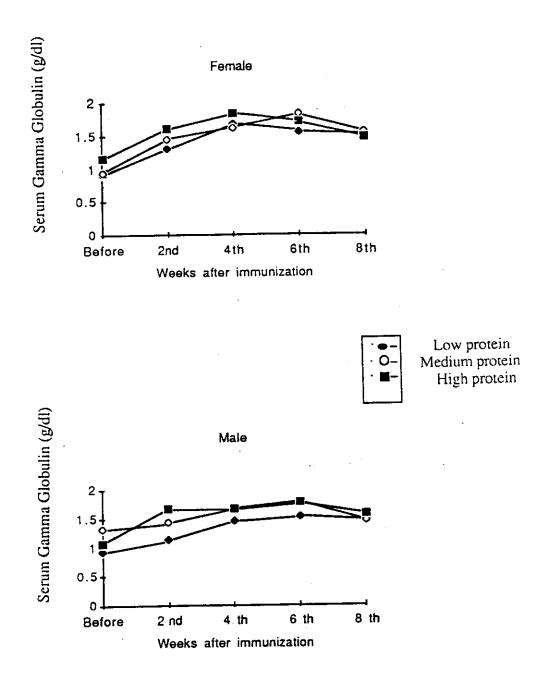


Fig (11) Serum Gamma globulin (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

4-1-3 Serum total thyroxine (T₄)

Data concerning the effect of dietary protein level and induced active thyroid immunity on serum total thyroxine concentration for both female and male Hubberd chicks along the experimental time before and after immunization are presented in table (8) and illustrated in figures 12 and 13.

Obtained results revealed that serum total thyroxine was significantly affected by dietary protein level, thyroglobulin immunization and time intervals (ANOVA table 9)

Protein level in chick's diet had siginficant effect on most experimental groups. Chicks fed low dietary protein had the lowest mean values of T_4 while those fed higher levels protein attained the highest values of serum T_4 . Medium protein diets fed to chicks were in between, similar results were obtained in most of female and male chicks. Serum T_4 avergad 1.46, 1.93 and 2.65 ugldl for female chicks fed low, medium and high dietary protein content, respectively. The corresponding values in males were 1.48, 1.78 and 1.99 ug/dl, respectively.

Induced thyroid immunity was found to affect serum T_4 significantly (Table 8). The highest mean values of T_4 were observed in chicks immunized with thyroglobulin. On the other hand, chicks of control or adjuvant injected groups attained approximately equal grand mean values in most of experimental chicks.

The overall mean values of serum T $_4$ of females were 1.67, 1.74 and 2.11 ugldl for control , adjuvant and thyroglobulin immunized group , respectively , the corresponding values in males were 1.66, 1.63 and 2.00 ugldl , respectively .

A consistent increase in serum total thyroxine was noted with the progess of time reaching its maximum values at the 4^{th} and 6^{th} weeks after

immunization , then decreased up to the end of the exprimental period . Mean values of T_4 were 1.86 $\mu g/dl$ before immunization and 1.81, 2.02, 1.86 and 1.74 ugldl at the three consecutive intervals for females . The corresponding values were in males 1.73, 1.71, 1.91, 1.74, and 1.68 ug/dl , respectively .

Significant interactions were found between active thyroid immunized groups and time intervals (ANOVA table 9). This result that the response of induced immunization differs according to the time to estimation after immunization.

The possibility that thyroid hormones play a role in antibody production was not ruled out (Mashaly et al., 1983). However, it could be suggested that the increasing levels of serum thyroid hormones as function of thyroglobulin immunization may be considered as indirect indication for antibody formation (Ibrahim et al., 1988).

The results of the present study reveald that serum level of thyroxine was increased as a function of immunization. This is in agreement with the results of Ibrahim and Premachandra., (1972).

The binding of T_4 by antithyroglobulin is a phenomenon described originally by Premachandra <u>et al</u>.,(1963) and confirmed by other investigators Mackenzie and Haibach; (1967) and Pogoriler <u>et al</u>.; (1971) Ibrahim and Premachandra, (1972) and Ibrahim <u>et al</u>.,(1988) during the last 20 years.

The increase in serum total thyroxine by inducing active thyroid immunity could be attributed to the extrabinding sites provided by thyroglobulin antibody molecules. It is worth mentioning that high concentrations of T_4 in immunized animals is not indicative of hyperfunctioning state of thyroid since the hormone bound to the antibody is physiologically inactive .

As thyroglobulin antibodies were demonastrated in non diseased and diseased humans and animals (Goudie . et al ., 1959; Roitt and Doniach, 1960; and Rose et al ., 1971). However, not all the naturally accuring thyroglobulin antibodies bind thyroxine (Pogoriler et al., 1971).

All these findings necesstiate the importance of critical evaluation of the thyroid function data in reference to possibly occurring antibodies.

As function of treatment, the increase in serum T_4 with the increasing levels of protein in diet, and decreasing with the lower levels were in agreement with Stewart and Washburn, (1984), who found that slightly deficient diet in protein, influence T_4 level in serum. These results may be explained by the marked reduction in thyroid growth which accompained by an apperent suppression of thyroidal activity. Protein difficiency in diet may result a decreased capacity of thyroid to concentrate iodide, so thyroid ability to synthesis hormones becomes inefficient.

The last mentioned results are in partial disagreement with Bryan and Jensen , (1989) who found that protein deficient diet had no influence on T_4 level in serum .

The decreasing serum T_4 levels with progress of age were in agreement with Falconer, (1966) who observed a linear decrease in secretion rate of chick's thyroid over the period of 14 to 100 days.

Table (8) Effect of dietary protein levels on serum Lotal thyroxine ug/dL (X± S.E) of Female and Male treated chicks at various time intervals before and after thyroglobulin immunization.

	SERUM THYROXINE, μg / dl								
Protein	C	Before	Weeks a	fter immı	inization				
level	Group	Deloie	2 nd	4 th	6 th	8 th			
FEMALE									
	Cont	1.62 <u>+</u> .05	1.49 <u>+</u> .09	1.49 <u>+</u> .09	1.35±.05	1.30 <u>+</u> .39			
Low	Adj	1.66 <u>+</u> .09	1.55 <u>+</u> .15	1.45±.47	1.50 <u>+</u> .57	1.43 <u>+</u> .09			
	Tg	1.58 <u>+</u> .14	1.93 <u>+</u> .43	2.23 <u>+</u> .13	1.93 <u>+</u> .15	1.63 <u>+</u> .24			
	Cont	1.89 <u>+</u> .10	1.77 <u>+</u> .13	1.58 <u>+</u> .17	1.53 <u>+</u> .44	1.52 <u>+</u> .09			
Medium	Adj	1.94 <u>+</u> .17	1.80 <u>+</u> .10	1.95 <u>+</u> .23	1.73 <u>+</u> .12	1.65 <u>+</u> .22			
	Tg	1.83 <u>+</u> .04	1.93 <u>+</u> .27	3.00 <u>+</u> .13	3.05 <u>+</u> .08	1.85 <u>±</u> .11			
	Cont	2.13 <u>+</u> .20			1.88 <u>+</u> .13				
High	Adj	1.89 <u>+</u> .22		i					
	Tg	2.24 <u>+</u> .16	2.77 <u>+</u> .34	2.68 <u>+</u> .31	2.00 <u>±</u> .20	2.03 <u>+</u> .11			
			MALE	,	T				
	Cont	1.50 <u>+</u> .07	1.30 <u>+</u> .85	1.13 <u>+</u> .09	1.19 <u>+</u> .04	1.46 <u>+</u> .15			
Low	Adj	1.60 <u>+</u> .07	1.48 <u>+</u> .07	1.02±.05	1.48 <u>+</u> .11	1.55 <u>+</u> .04			
	Tg	1.52±.31	1.62 <u>+</u> .40	1.97±.21	1.95±.25	1.70 <u>±</u> .23			
	Cont	1.69 <u>+</u> .07	1.65±.32	1.35 <u>+</u> .22	1.60 <u>+</u> .15	1.57 <u>+</u> .08			
Medium	Adj	1.80 <u>+</u> .07	1.72 <u>+</u> .05	1.80 <u>+</u> .14	1.68 <u>+</u> .11	1.60 <u>+</u> .04			
	Tg	1.87 <u>+</u> .23	1.95 <u>+</u> .17	2.46 <u>+</u> .09	1.91 <u>±</u> .03	2.06 <u>+</u> .13			
	Cont	1.96 <u>+</u> .01	1.81 <u>+</u> .15	1.85 <u>+</u> .17	1.91 <u>±</u> .06	1.73 <u>+</u> .73			
High	Adj	1.99 <u>+</u> .15	1.93 <u>+</u> .32	2.01 <u>+</u> .18	1.78 <u>+</u> .15	1.86 <u>+</u> .31			
	Tg	1.79 <u>+</u> .26	1.95 <u>+</u> .06	2.70±.45	2.21 <u>+</u> .06	2.45 <u>+</u> .11			

N.B Cont = control group, Adj = adjuvant injected group.

Tg = Thyroglobulin immunized group

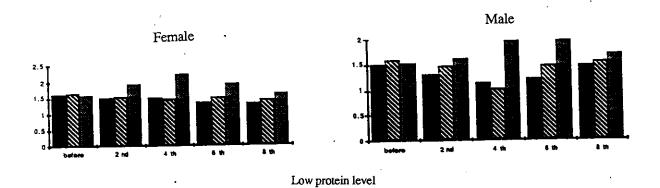
Table (9) Analysis of variance for the Data represented in table (8)

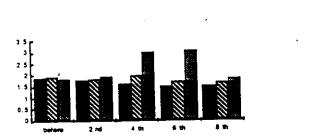
Source of variation	D.F	Sum of squares	Mean squares	F
Sex (S)	1	.288	.288	1.764
Protein level (p)	· 2	7.589	3.794	23.229***
Immunization (M)	2	11.675	5.837	35.733***
Interval (I)	4	8.539	2.134	13.069***
SxP	2	.379	.189	1.161
SxM	2	.522	.261	1.601
SxI	4	.418	.104	.640
PxM	4	.332	.083	.509
PxI	8	1.998	.249	1.529
MxI	8	5.927	.740	4.536**
Remainder	232	37.900	1.633	

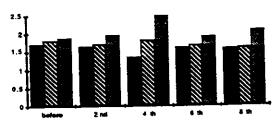
^{*} P < 0.05

^{**} P < 0.01

^{***} P < 0.001







Medium protein level

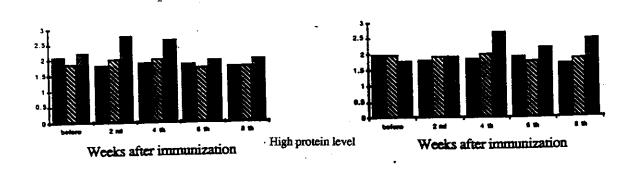


Fig (12) Serum total thyroxine (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.

-	I	Cont
4	2	Adj
.l		Tg

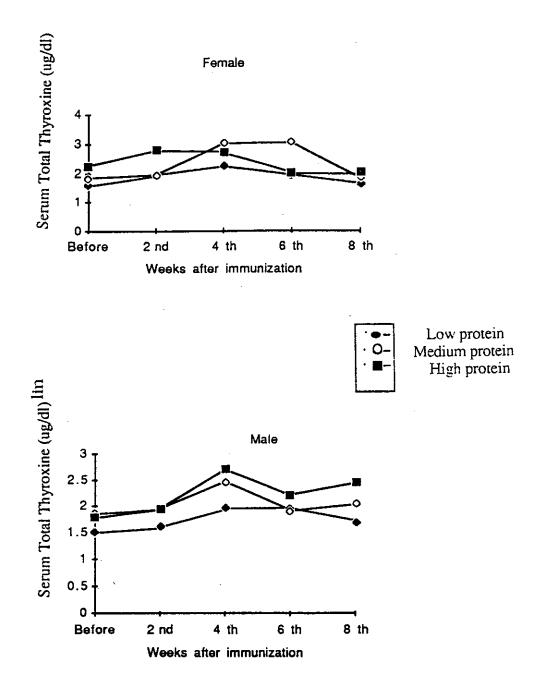


Fig (13)Serum Lotal thyroxine (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

4.1-4 Serum Triiodothyronine (T₃)

Data concerning the effect of dietary protein and thyroglobulin immunization on serum triiodothyronine concentration in female and male Hubbard chicks are presented in table (10) and illustrated in figures 14 and 15

Significant difference due to the protein level in diet and time intervals before and after immunization were observed (ANOVA table 11).

Little variations in serum T_3 mean values as effect of feeding the chicks different levels of protein in diet were noted. Females grand means amounted to 137.60, 140.60 and 149.54 ng / dl for chicks fed low, medium and high dietary protein levels, resecptively, the corresponding values for males were 142.28, 134.11 and 154.80 ng/dl, respectively.

Variations due to time intervals showed inconsistent trend in the overall means of the experimental group of chicks. Serum T₃ of famales were 139.44 ng/dl before immunization and 139.71, 149,63, 139,3 and 143.9 ng/dl at the 2nd, 4th, 6th and 8th week after immunization, respectively, while the corresponding values in males were 139.07 ng/dl before immuization and 140.08, 135.92, 151.26 and 145.75 ng/dl thereafter, respectively.

Significant interaction was found between time intervals and immunization (ANOVA table 11). This indicates that immunization affect serum T_3 in different manner with each time interval after immunization.

The role of thyriod gland in influencing humoral immunty either in avian or mammalin species is not well established (Wick et al., 1985).

Under the present experimental conditions, estimated T₃ in serum was affected by protein level in diet and time intervals after immunization. Little

increase, was noted in thyroglobuin immunized groups which was indicative to antibody formation. This may explained by the extrabinding sites provided by thyroglobulin immunization.

It is worth mentioning that the determination of serum triiodothyronine as an index of antithyroglobulin antibodies is indirect method and it may lack sensitivity in cases of low antibody titre (Ibrahim and Premachandra, 1972).

It is of a great importance to mention that chick thyroiditis are usually accompained by the presense of auto antibodies against thyroid hormones (Chan et al., 1989). However, the inconsistency between T_4 and T_3 binding to antibody indicates that T_4 is most potent in binding thyroglobulin antibodies than T_3 (Odette, 1986 and Ibrahim et al., 1988).

Concerning the effect of dietary protein on serum T_3 , Stewart and Washburn, (1984) were inagreement with the present experimental results that feeding diet slightly deficient in protein had little effect on T_3 concerntration in serum. However, Alster and Carew, (1984) and Keagy et al., (1987) observed that plasma T_3 increased when chicks fed protein deficient diet.

Evidence to clarify the exact role of the thyroid hormones in the metabolism of protein deficient chick remains incomplete (Bryan and Jensen, 1989). However, it could be recommended to carry out additional studies to find the relationships between thyroid hormones and the naturally or stimulated auto antibodies formed against the thyroid hormones and the common disorders affecting the rate of metabolism and lymphocytic filtration of the gland.

Table (10) Effect of dietary protein levels on serum Triiodothyronine ng/dI (X±S.E) of Female and Male treated chicks at various time intervals before and after thyroglobulin immunization.

	SERUM TRIIODOTHYRONINE,ng/dl							
Protein	Group	Before	Weeks	after imm	unization			
level	Group	Belore	2 nd	4 th	6 th	8 th		
- 	- . <u>-</u>		FEMAI	Æ				
	Cont	135.0 <u>±</u> 10	118.4±17	136.2 <u>+</u> 4	136.5 <u>+</u> 4	129.2 <u>+</u> 4		
Low	Adj	131.0 <u>+</u> 16	161.8 <u>+</u> 6	151.8 <u>+</u> 10	121.0 <u>+</u> 13	166.0 <u>+</u> 17		
	Tg	135.3 <u>+</u> 14	141.7 <u>+</u> 29	132.0 <u>+</u> 5	139.1 <u>+</u> 13	129.0 <u>+</u> 8		
;	Cont	126.6 <u>+</u> 11	123.5 <u>+</u> 28	149.9 <u>+</u> 6	126.8 <u>+</u> 6	147.3 <u>+</u> 7		
Medium	Adj	139.9 <u>+</u> 8	133.3 <u>+</u> 28	166.7 <u>+</u> 24	119.8 <u>±</u> 24	118.0 <u>+</u> 6		
	Tg	136.8 <u>+</u> 16	173.0 <u>+</u> 6	158.1 <u>+</u> 20	131.8 <u>+</u> 24	149.7 <u>+</u> 5		
	Cont	135.5 <u>+</u> 12	132.2 <u>+</u> 5	141.2 <u>+</u> 16	147.2 <u>+</u> 7	159.4 <u>+</u> 13		
High	Adj	146.3±14	127.8 <u>+</u> 23	135.8 <u>+</u> 16	168.3 <u>+</u> 11	155.0 <u>+</u> 15		
	Tg	168.7 <u>+</u> 16	145.7 <u>+</u> 22	175.0 <u>+</u> 28	163.3 <u>+</u> 4	141.8 <u>+</u> 21		
			MAI	Œ				
	Cont	143.9 <u>+</u> 10	121.7 <u>+</u> 8	136.2 <u>+</u> 23	151.0 <u>+</u> 17	157.7 <u>+</u> 11		
Low	Adj	133.6 <u>+</u> 16	160.2 <u>±</u> 13	150.0 <u>+</u> 8	126.0 <u>+</u> 33	133.6 <u>+</u> 10		
	Tg	128.3 <u>+</u> 20	127.5 <u>+</u> 8	154.4 <u>+</u> 15	166.2 <u>+</u> 22	139.0 <u>+</u> 10		
	Cont	112.5 <u>+</u> 16	123.3 <u>+</u> 8	121.8 <u>+</u> 9	136.3 <u>±</u> 12	152.5 <u>+</u> 19		
Medium	Adj	146.8 <u>+</u> 7	126.8 <u>±</u> 15	130.3 <u>+</u> 7	127.3 <u>+</u> 31	119.0 <u>+</u> 13		
	Tg	140.0 <u>+</u> 25	131.8 <u>+</u> 4	151.1 <u>+</u> 13	157.2 <u>+</u> 21	133.3 <u>+</u> 3		
High	Cont	153.9 <u>+</u> 8	153.7 <u>+</u> 9	164.3 <u>±</u> 10	139.7 <u>+</u> 11	165.7 <u>+</u> 6		
	Adj	136.3 <u>±</u> 12	135.0 <u>+</u> 28	180.3 <u>+</u> 18	141.0 <u>+</u> 12	146.0 <u>+</u> 10		
	Tg	165.1 <u>+</u> 16	143.3 <u>+</u> 22	171.8 <u>+</u> 16	161.0 <u>+</u> 16	165.0 <u>+</u> 5		

N.B Cont = control group, Adj = adjuvant injected group.

Tg = Thyroglobulin immunized group

Table (11) Analysis of variance for the Data represented in table (10)

Source of variation	D.F	Sum of squares	Mean squares	F
Sex (S)	1	498.032	498.032	0.664
Protein level (p)	. 2	4892.605	2446.302	3.262*
Immunization (M)	2	2932.999	1466.499	1.955
Interval (I)	4	15799.188	3949.797	5.267***
SxP	2	56.429	28.214	.038
SxM	2	1375.609	687.804	.917
SxI	4	3789.498	947.374	1.263
PxM	4	1557.551	389.387	.519
PxI	8	5595.262	699.407	.933
MxI	8	14811.496	1851.437	2.467**
Remainder	232	1738 7 .345	749.946	

^{*} P < 0.05

^{**} P < 0.01

^{***} P < 0.001

Female

Weeks after immunization

Male

Weeks after immunization

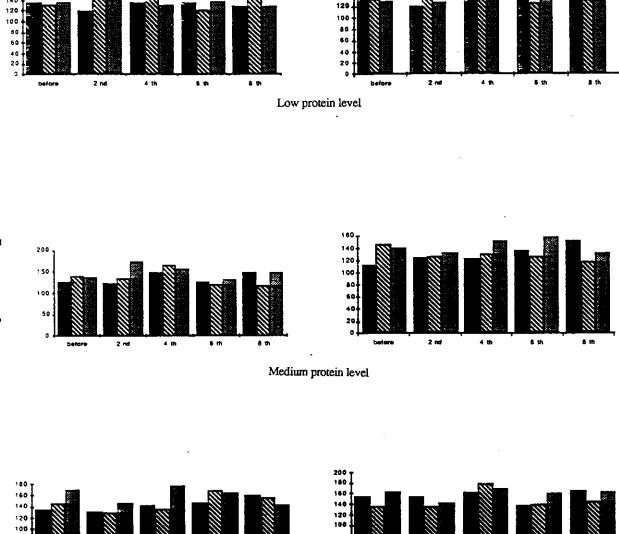


Fig (14) Serum triiodo-thyronine (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.

High protein level

	Cont
2	Adj
9 - 962	Tg

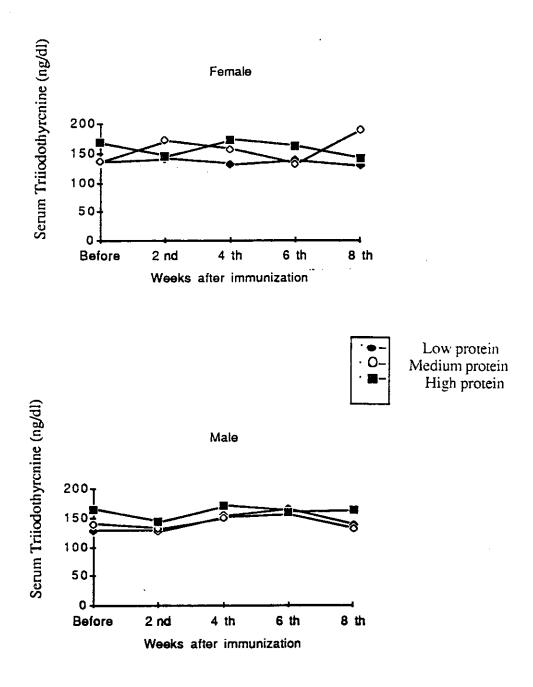


Fig (15) Serum Triiodo thyronine (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

4-2 : Effect of dietary protein level and thyroglobulin immunization on some metabolic blood parameters : -

4-2-1- Serum total proteins :-

The average serum total protein values of all group studied are presented in table (12) and illustrated in figures 16 and 17. Significant differences in serum total proteins levels due to dietary protein levels, immunization and time intervals were obtained (ANOVA table 13).

Chicks fed relatively low dietary protein had the lowest values of serum total protein, while those fed high protein level in diet had the highest serum protein mean values. This trend was clear in both female and male chicks. Serum protein grand mean values were 3.43, 3.98 and 4.6 g/dl for female chicks fed low, medium and high protein level in diet, while the corresponding values were 3.66, 4.22 and 4.67 g/dl in males, respectively.

Approximate values in serum total protein were obtained in both the control and adjuvant injected chicks, while it was little higher in thyroglobulin immunized groups. Serum total proteins grand means were 3.97, 3.90 and 4.23 gldl for control, adjuvant and immunized female chicks, respectively, The corresponding values were 4.2, 4.18 and 4.48 g/dl in males, respectively.

With the progress of time after immunization, a gradual and consistent increase in serum total proteins was observed for both female and male chicks. Serum total proteins grand means were (3.56 gldl) before immunizing female chicks and 4.12, 4.36, 4.38 and 4.49 g/dl at 2nd, 4th, 6th and 8th weeks there after. The corresponding values were 3.63, 4.16, 4.24, 4.41 and 4.70 g/dl in male chicks, respectively.

Significant effects were observed due to the interaction between immunization and sex. This may lead to conclude that immunization effect differed signficantly according to the chicks sex and was modified as time after

immunization passed. A significant interaction effect between dietary protein levels and time intervals was also noted.

4-2-2 Serum Albumin:

Data listed in table (14) present serum albumin mean values as affected by dietary protein level and immumization at various time in ntervals in both female and male experimental Hubbard chicks, and illustrated in figures 18 and 19.

Obtained results reveal that Serum albumin was significantly affected by protein level in diet and time intervals. While no significant effects due to either chick's sex or immunization were observed (ANOVA table 15).

Chicks fed low level of protein in diet had low mean values of serum albumin, Those fed diet containing high protein level had the higherst albumin values than those fed medium diet. Serum albumin over all mean values were 1.49, 1.62 and 1.78 g/dl for female chicks fed low, medium and high levels of protein in diet, respectively. The corresponding values in males were 1.50, 1.62 and 1.75 g/dl, respectively.

Serum albumin mean values were increased with the progess of time , reaching its highest values at the $8^{\rm th}$ week after immunization . Albumin grand mean values were 1.56, 1.59, 1.64, 1.69 and 1.71 g/dl before and at the $2^{\rm nd}$, $4^{\rm th}$, $6^{\rm th}$ and $8^{\rm th}$ week after immunization of females. The corresponding values in males were 1.59, 1.58 1.59, 1.64 and 1.80 g/dl respectively

Significant effect was obtained due to the interaction between sex and dietary protein level indicates that dietary protein affects serum albumin levels in females in different mannars than males.

4-2-3 Serum globulins:

Data concerning the effect of dietary protein level and active thyroid immunization on female and male chicks on serum globulins are presented in table (16) and illustrated in figures 20 and 21.

Significant difference in serum globulins due to sex, dietary protein level, immunization and time intevals were observed and due to interactions between sex ximmunization and protein level x time interval (ANOVA table 17).

Regardless the effect of any other factor, female chicks had higher grand mean (2.95 g/dl) of serum globulins than males (2.63 g/dl).

The effect of dietary protein on serum globulin levels was pronounced in all experemintal groups, higher values were associated with higher dietary protein levels and those chicks fed low protein diet showed relatively low levels of serum globulins.

The grand mean values of female serum globulins were 2.42, 2.68 and 2.78 g/dl for chicks fed low, medium and high levels of protein in diet. while the corresponding values in males were 2.47, 2.65 and 2.75 g/dl , respectively.

Thyroglobulin immunization significantly (P < 0.05) affected serum globulins, It's grand means were higher in both female and male immunized groups than the control . Serum globulins mean values for females were 2.60, 2.59 and 2.91 g / dI $\,$ for control , adjuvant and thyroglobulin immunized groups , respectively , while the corresponding values in males were 2.51, 2.70 and 2.79 g / dI $\,$, respectively .

4-2-4 Albumin / globulin Ratio (A/G ratio):

The effect of dietary protein, active thyroid immunity, chick's sex and time intervals on albumin / globulin ratio are presented in table 18 and illustrated in figures 22 and 23.

Significant difference in A/G values were observed due to sex, time intervals and the interaction between protein level and time intervals and between sex and immunization (ANOVA table 19).

Female chicks had mostly higher A/G mean values than males which averaged (0.664 and 0.613) for female and male chicks, respectively.

Decreased A/G values were observed with the progress of time , it averaged 0.69 , 0.63 , .0.60 , 0.57 and 0.57 before immunization and after $2^{\rm nd}$, $4^{\rm th}$, $6^{\rm th}$ and $8^{\rm th}$ weeks after in female chicks while the corresponding values in males were 0.67 , 0.58 , 0.56 . 0.55 and 0.58 , respectively .

To achieve optimum broiler growth, diet must provide sufficient levels of essential amino acids to meet the demands of metabolic processes. The crude protein components of a diet used, have to provide a balanced pattern of amino acid to promote optimal energy and protein intake (Bryan and Jensen, 1989).

Under the present experimental conditions, serum total protein, albumin and globulins levels were directly affected by the level of protein in diet. A lower values of the mentioned parameters were always associated with feeding the experimental chicks low levels of protein in diet. However, higher values of serum total protein, albumin and globulins were associated with higher dietary protein levels.

As it is already reviewed, plasma protein is considerd to be an indicator for the status of nitrogen in diet. However, the previous results may be attributed to the amount of avilable nitrogen in diet, thus the inadquate supply of nitrogen or amino acids in low protein level diet in turn affect the total protein level in serum and generally decreases the rate of albumin synthesis.

Results obtained by (Mukhtar et al., 1985; Heibashy, 1990 and Payne et al., 1990), were in agreement with the present results indicating that serum total protein and albumin were both directly related to the level of protein in diet.

Low protein level in diet may appear in case of hypoproteinemia, while higher levels than needed for the different physiological processes may cause a case of hyperproteinemia to the growing chicks (Micheal and David, 1983).

As afunction of thyroglobulin immunization, Significant effects obtained in both serum total protein and globulins in all the experimental dietary protein levels used, this may be due to the intensive continuous need for serum proteins mainly globulins durning which the antigen was processed and cellular proliferation and differentiation accurd, thus circulating antibodies could be detected. However, a partial agreements with the present results were supported by (Azeez and Paily, 1985) who found a reduction in serum protein values in low antibody titre group of chicks vaccinated with Newcastle disease indicated an immune suppressive effect.

Increased levels of serum total protein and globulin in the immunized groups of chicks with the progress of time ofter immunization were in ageement with other investigators in different species. Ibrahim, (1973) in rats, Abd EL Moneim, (1980) in rabbits, and Oyeijd et al., (1985) in chickens.

Concerning the flactuating values of serum albumin and A/G ratio as effect of immunization, Similar results were reported by Odette, (1986). However, Ibrahim, (1973) observed no change in serum total protein, albumin and globulin as effect of thyroglobulin immunization. Such inconsietency may be attributed to the difference in the species of the animal used.

Whereas albumin abnormalties are largely restricted to decreased levels in serum in association to protein deficiency or protein losting state (Peter and Gorevic, 1983, the major components of serum globulins abnormalities are largerly restricted to defence mechanisms against infectian and inflammations. However, gamma globulin is an excellent screen for the status of humoral, T cell-mediated immunity. This may explain effect of thyroglobulin immunization on serum globulins.

It could be recommended that diet should be formulated to avoid both protein deficiency or surplus to achieve efficient protein utilization.

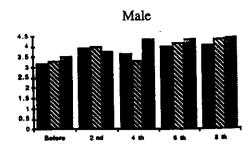
Table (12) Effect of dietary protein levels on serum Total protein g/dI (X± S.E) of Female and Male treated chicks at various time intervals before and after thyroglobulin immunization.

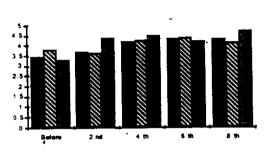
	SERUM TOTAL PROTIEN, g/dI								
Protein Group Before Weeks after immunization									
level	Group	Deloi e	2 nd	4 th	6 th	8 th			
	FEMALE								
	Cont	3.10 <u>+</u> .12	3.89 <u>+</u> .12	4.03 <u>±</u> .20	3.84 <u>+</u> .15	4.15 <u>+</u> .11			
Low	Adj	3.19 <u>+</u> .14	3.53 <u>+</u> .15	3.97 <u>+</u> .12	4.04 <u>+</u> .22	4.06 <u>±</u> .13			
·	Tg	3.01±.12	3.95±.18	4.06 <u>+</u> .14	4.14 <u>+</u> .24	4.02 <u>+</u> .20			
	Cont	3.49 <u>+</u> .18	3.69 <u>+</u> .18	4.18 <u>+</u> .19	4.32 <u>+</u> .16	4.30 <u>+</u> .15			
Medium	Adj	3.79 <u>+</u> .21	3.62 <u>±</u> .14	4.24 <u>+</u> .17	4.37 <u>+</u> .14	4.14 <u>+</u> .10			
	Tg	3.29 <u>+</u> .15	4.35 <u>+</u> 17	4.48 <u>+</u> .13	4.21 <u>+</u> .15	4.71 <u>+</u> .15			
	Cont	3.96 <u>+</u> .13	4.48 <u>+</u> .17	4.84 <u>±</u> .14	4.88 <u>+</u> .08	5.35 <u>+</u> .09			
High	Adj	4.29 <u>+</u> .17	4.45 <u>+</u> .18	4.63 <u>+</u> .15	4.77±.21	4.87 <u>+</u> .12			
	Tg	3.97 <u>+</u> .17	4.39 <u>+</u> .13	4.89 <u>+</u> .12	4.91 <u>+</u> .17	4.88 <u>+</u> .15			
				MALE					
	Cont	3.19 <u>+</u> .13	3.92 <u>+</u> .08	3.62 <u>+</u> .19	4.01 <u>+</u> .18	4.05 <u>+</u> .14			
Low	Adj	3.30 <u>+</u> .14	4.00±.18	3.30 <u>+</u> .14	4.15 <u>+</u> .30	4.30 <u>+</u> .20			
	Tg	3.50 <u>+</u> .02	3.75 <u>±</u> .16	4.34 <u>+</u> .04	4.30 <u>+</u> .17	4.37±.17			
	Cont	3.60±.14	4.27 <u>+</u> .18	4.40±.13	4.25 <u>+</u> .19	4.38 <u>+</u> .18			
Medium	Adj	3.35 <u>+</u> .15	4.32 <u>+</u> .07	4.34 <u>+</u> .16	4.46 <u>+</u> .09	4.41 <u>+</u> .14			
	Tg	3.84±.21	4.17 <u>+</u> .18	4.11±.17	4.56 <u>+</u> .09	4.77 <u>+</u> .12			
	Cont	4.28 <u>+</u> .11	3.82 <u>+</u> .13	4.63 <u>+</u> .18	4.85 <u>+</u> .14	5.73 <u>+</u> .11			
High	Adj	3.86±.10	4.89 <u>+</u> .13	4.68 <u>+</u> .05	4.60 <u>+</u> .08	4.83 <u>+</u> .11			
	Tg	4.22 <u>+</u> .15	4.53 <u>+</u> .20	4.82 <u>+</u> .07	5.13 <u>+</u> .08	5.08 <u>+</u> .23			

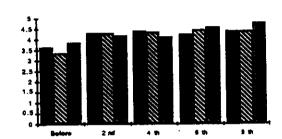
N.B Cont = control group, Adj = adjuvant injected group. Tg = Thyroglobulin immunized group

Table (13) Analysis of variance for data represented in table (12)

Source of variation	D.F	Sum of squares	Mean squares	F
Sex (S)	1	.591	.591	2.942
Protein level (p)	2	29.914	14.957	74.401***
Immunization (M)	2	1.143	.571	2.844*
Interval (I)	4	75.762	18.940	94.215***
SxP	2	.370	.185	.922
SxM	2	3.040	1.520	7.562***
SxI	4	1.256	.314	1.563
PxM	4	.8250	.206	1.026
PxI	8	4.443	.555	2.763**
MxI	8	.739	.092	.460
Remainder	637	128.060	.201	







Medium protein level

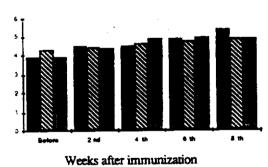




Fig (16) Serum Total proteins (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.

Serum Total proteins g / dL

Adj Tg

Cont

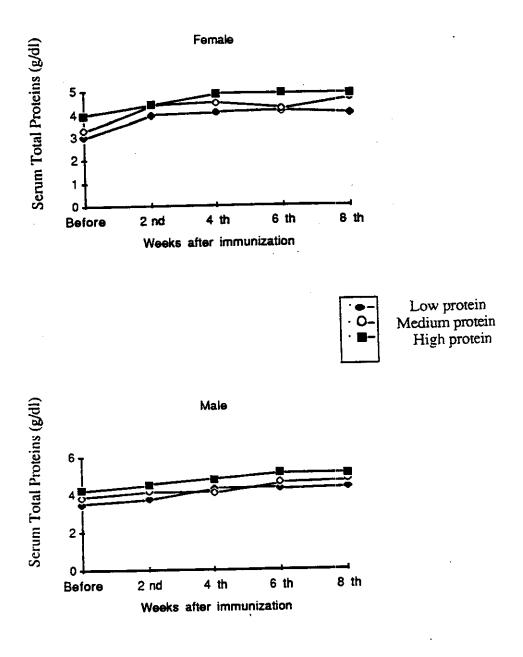


Fig (17) Serum Total proteins (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

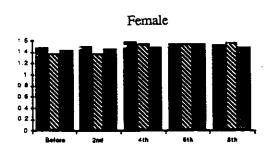
Table (14)Effect of ditary protein levels on serum Albumin g/dl ($\overline{X} \pm S-E$) of Female and Male treated chicks at various time intervals before and after thyroglobulin immunization.

	SERUM ALBUMIN, g/dl							
Protein	Group	Before	Weeks a	fter immı	inization			
level	Group	Delore	2 nd	4 th	6 th	8 th		
			FEMALE	C				
	Cont	1.48 <u>+</u> .04	1.51 <u>+</u> .06	4.59 <u>+</u> .07	1.54 <u>+</u> .07	1.52 <u>+</u> .04		
Low	Adj	1.37±.04	1.39±.07	1.55 <u>±</u> .06	1.55 <u>+</u> .07	1.57 <u>+</u> .07		
	Tg	1.45 <u>+</u> .03	1047 <u>+</u> .05	1.48 <u>+</u> .07	1.54 <u>±</u> .05	1.48 <u>+</u> .07		
	Cont	1.57 <u>+</u> .09	1.59 <u>+</u> .06	1.66 <u>+</u> .08	1.63 <u>+</u> .06	1.65 <u>+</u> .07		
Medium	Adj	1.65 <u>+</u> .09	1.56 <u>±</u> .07	1.62±.06	1.55±.08	1.64 <u>+</u> .05		
•	Tg	1.51 <u>+</u> .06	1.65 <u>+</u> .06	1.63 <u>+</u> .07	1.57 <u>+</u> .04	1.82 <u>+</u> .08		
	Cont	1.64 <u>+</u> .06	1.64 <u>+</u> .08	1.77 <u>+</u> .08	1.82 <u>+</u> .03	1.81 <u>+</u> .05		
High	Adj	1.76 <u>+</u> .05	1.64 <u>+</u> .06	1.70 <u>+</u> .06	1.81 <u>+</u> .07	1.94 <u>+</u> .06		
	Tg	1.61 <u>+</u> .06	1.74±.07	1.77 <u>±</u> .07	1.74 <u>+</u> .08	1.97 <u>+</u> .04		
			MALE					
:	Cont	1.42 <u>+</u> .05	1.51±.07	1.42 <u>+</u> .09	1.47 <u>+</u> .06	1.49 <u>+</u> .04		
Low	Adj	1.50 <u>+</u> .08	1.59 <u>+</u> .06	1.51±.03	1.40 <u>+</u> .06	1.54 <u>+</u> .09		
	Tg	1.54 <u>+</u> .04	1.54 <u>+</u> .04	1.58 <u>+</u> .05	1.52 <u>+</u> .05	1.54 <u>+</u> .08		
	Cont	1.62 <u>+</u> .09	1.58 <u>+</u> .02	1.64 <u>+</u> .05	1.69 <u>+</u> .07	1.66±.07		
Medium	Adj	1.59 <u>+</u> .09	1.49±.06	1.56±.04	1.63±.07	1.72 <u>+</u> .23		
	Tg	1.58 <u>+</u> .08	1.53 <u>±</u> .03	1.78 <u>±</u> .06	1.63 <u>+</u> .08	1.72 <u>+</u> .23		
	Cont	1.64 <u>+</u> .08	1.61 <u>+</u> .07	1.66 <u>+</u> ;.05	1.81±.04	1.86 <u>+</u> .09		
High	Adj	1.79 <u>+</u> .09	1.69 <u>+</u> .04	1.60 <u>+</u> .07	1.75±.05	1.91 <u>+</u> .06		
	Tg	1.78±.08	1.69. <u>+</u> .03	1.83 <u>±</u> .05	1.87±.05	1.82 <u>+</u> .09		

Table (15) Analysis of variance for Data represented in table (14)

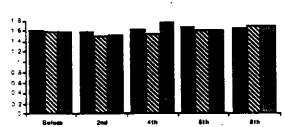
Source of variation	D.F	Sum of squares	Mean squares	F
Sex (S)	1	.005	.005	.150
Protein level (p)	2	4.003	2.021	53.524***
Immunization (M)	2	.190	.095	2.520
Interval (I)	4	.755	.188	5.002***
SxP	2	.221	.110	2.927*
SxM	2	.067	.033	.895
SxI	4	.164	.041	1.087
PxM	4	.048	.012	.322
PxI	8	.389	.048	1.288
MxI	8	.507	.063	1.680
Remainder	637	24.059	.037	

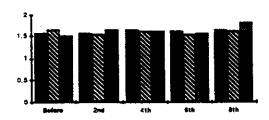




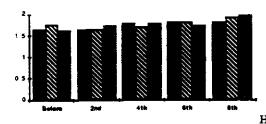


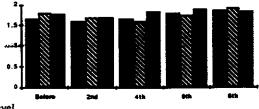






Medium protein level





Weeks after immunization

High protein level Weeks after immunization

Fig (18) Serum Albumin (means) of Fernale and Male chicks at low, medium and high levels dietary protein before and after immunization.

Cont

Adj

Tg

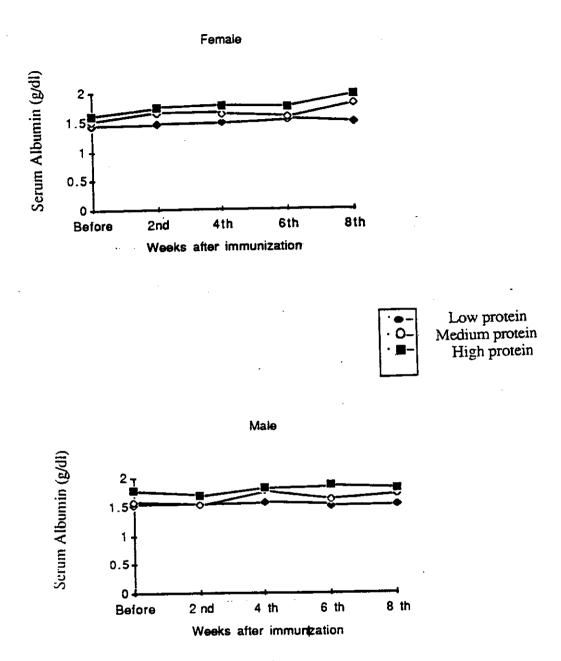


Fig (19) Serum Albumin (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

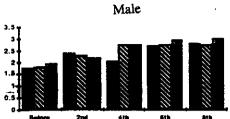
Table (16) Effects of dietary protein levels on serum globulins g/dl (x± S.E) of female and Male treated chicks at various time intervals before and after thyroglobulin immunization

	SERUM GLOBULINS g/dl						
Protein	· GIVIU DEIVIE						
level		•	2 nd	4 th	6 th	8 th	
			FE	MALE			
	Cont	1.72± .08	2.38 <u>+</u> .09	2.74 <u>+</u> .13	2.92 <u>+</u> .11	2.78 <u>+</u> .08	
Low	Ad j	1.82 <u>+</u> .11	2.22 <u>+</u> .17	2.47 <u>+</u> .14	2.50±.11	2.80 <u>+</u> .07	
:	Tg	1.85±.09	2.49 <u>+</u> .09	2.58 <u>+</u> .17	2.60 <u>+</u> .26	2.94 <u>+</u> .10	
	Cont	1.92 <u>+</u> .10	2.23 <u>+</u> .17	2.61 <u>+</u> .17	2.79 <u>+</u> .12	2.79 <u>+</u> .09	
Medium	Adj	1.97 <u>+</u> .14	2.23±.03	2.61 <u>±</u> .10	2.84±.13	2.91 <u>+</u> .08	
i	Tg	2.71 <u>+</u> .13	2.67 <u>+</u> .08	2.73 <u>+</u> .18	2.93 <u>+</u> .15	2.99 <u>+</u> .08	
	Cont	2.32 <u>±</u> .08	2.66±.19	2.97 <u>+</u> .15	2.87+10	2.94 <u>+</u> .50	
High	Adj	2.21±.15	2.81±.10	2.91 <u>+</u> .13	2.85+10	2.94 <u>+</u> .12	
	Tg	2.17 <u>+</u> .13	2.64 <u>+</u> .15	2.94 <u>±</u> .13	2.97+15	3.14 <u>+</u> .18	
				MALE			
ĺ	Cont	1.77 <u>+</u> .08	2.41 <u>+</u> .12	2.08 <u>+</u> .15	2.72 <u>+</u> .17	2.85 <u>+</u> .18	
Low	Adj	1.81 <u>+</u> .07	2.31±.17	2.78±.05	2.75±.13	2.74 <u>+</u> .11	
	Tg	1.97 <u>+</u> .09	2.24 <u>+</u> .12	2.75 <u>+</u> .17	2.99 <u>+</u> .24	3.01 <u>+</u> .18	
	Cont	1.87±.03	2.60 <u>+</u> .18	2.75±.13	2.75 <u>+</u> .14	2.70 <u>±</u> .13	
Medium	Adj	2.89±.26	2.74 <u>+</u> .13	2.78 <u>+</u> .14	2.83 <u>+</u> .19	2.83±.10	
	Tg	2.00±.09	2.59 <u>+</u> .09	2.55 <u>+</u> .07	2.92 <u>+</u> .37	3.04 <u>+</u> .10	
	Cont	2.19 <u>+</u> .10	2.67±.09	2.90 <u>+</u> .17	2.84 <u>±</u> .13	2.58 <u>+</u> .09	
High	Adj	2.57 <u>+</u> .18	2.71 <u>+</u> .15	2.88±.05	2.96 <u>+</u> .07	2.86 <u>+</u> .09	
	Tg	2.28 <u>+</u> .07		3.08 <u>+</u> .06	3.18±.05	3.09±.09	

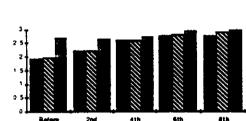
Table (17) Analysis of variance for the Data represented in Table (15)

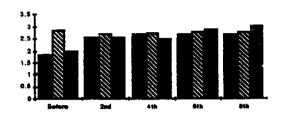
Source of variation	D.F	Sum of squares	Mean squares	F
Sex (S)	1	1.463	1.463	6.890**
Protein level (p)	2	8.025	4.012	18.887***
Immunization (M)	2	.931	.465	2.193*
Interval (I)	4	66.603	16.650	78.367***
SxP	2	.237	.118	.558
SxM	2	2.036	1.018	4.792**
SxI	4	.765	.191	.900
PxM	4	.834	.208	.982
PxI	8	3.515	.439	2.068*
MxI	8	1.734	.216	1.020
Remainder	637	135.344	.212	



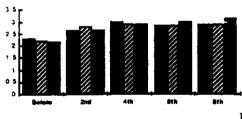


Serum Globulins g / db





Medium protein level



2.5 2.5 1.5 1.5 0.5

Weeks after immunization

Weeks after immunization

High protein level

Fig (20) Serum globulins (means) of Fernale and Male chicks at low, medium and high levels dietary protein before and after immunization.



Cont

Adj

Tg

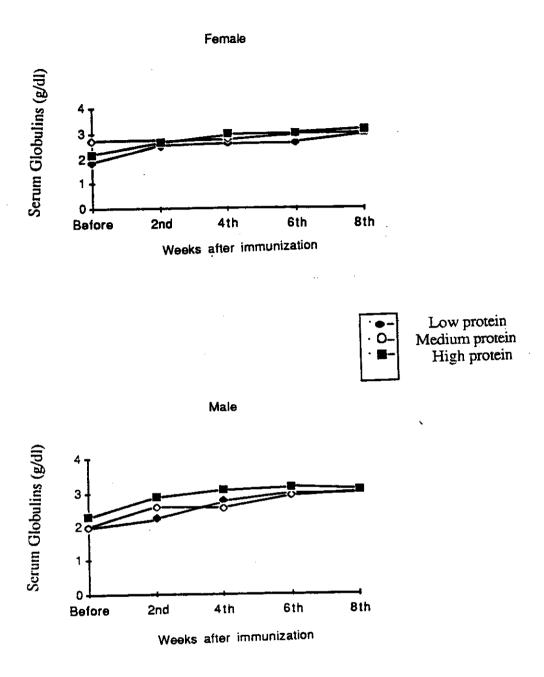


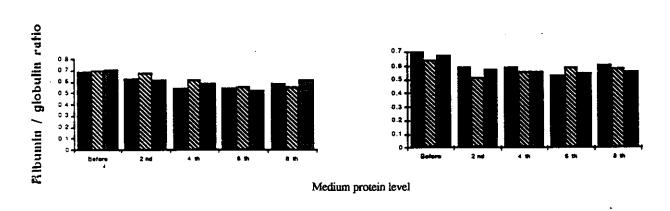
Fig (19) Serum Globulins (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

Table (18) Effect of dietary protein levels on albumin /globulin ratio ($\bar{x} \pm S \cdot E$) of Female and Male treated chicks: at various time intervals before and ofter thyroglobulin immunization

ALBUMIN / GLOBULIN RATIO									
Protein		Before		ter immu					
level	Group	Belore	2 nd	4 th	6 th	8 th			
	FEMALE								
	Cont	0.64 <u>+</u> .03	0.63 <u>±</u> .03	0.58 <u>+</u> .02	0.53 <u>+</u> .03	0.52 <u>+</u> .01			
Low	Adj	0.66 <u>+</u> .09	0.59 <u>+</u> .04	0.58±.06	0.54 <u>+</u> .06	0.56 <u>+</u> .05			
	Tg	0.68 <u>+</u> .04	0.60 <u>+</u> .04	0.60 <u>±</u> .03	0.54 <u>+</u> .05	0.57 <u>+</u> .02			
	Cont	0.69±.02	0.63 <u>+</u> .02		0.54 <u>±</u> .02				
Medium	Adj	0.70 <u>+</u> .08	0.68±.01	0.61 <u>+</u> .04	0.56±.02	0.56 <u>+</u> .01			
,	Tg	0.71±.03	0.61 <u>+</u> .05	0.58 <u>+</u> .03	0.53 <u>+</u> .04	0.61±.02			
	Cont	0.70 <u>+</u> .01	0.67 <u>+</u> .06	0.53 <u>+</u> .03	0.60 <u>+</u> .03	0.61 <u>±</u> .02			
High	Adj	0.73±.06	0.66 <u>+</u> .04	0.58 <u>+</u> .04	0.64 <u>+</u> .02	0.57±.03			
	Tg	0.68±.05	0.64 <u>+</u> .03	0.60 <u>+</u> .02	0.60±.03	0.59±.02			
				MALE	 	1			
	Cont	0.66±.01	0.58±.03	0.56 <u>+</u> .04	0.55±.02	0.58 <u>+</u> .06			
Low	Adj	0.63 <u>+</u> .09	0.56±.05	0.55±.02	0.56±.03	0.58 <u>+</u> .01			
	Tg	0.68±.10	0.53 <u>+</u> .01	0.54±.04	0.54±.04	0.50 <u>+</u> .05			
	Cont	0.70±.03	0.59 <u>+</u> .01	0.59±.02	0.53±.03	0.61 <u>+</u> .05			
Medium	Adj	0.64±.09	0.51±.03	0.56±.04	0.59±.09	0.58 <u>+</u> .03			
	Tg	0.68 <u>+</u> .10	0.57 <u>+</u> .05	0.56 <u>+</u> .02	0.55 <u>+</u> .02	0.56 <u>+</u> .03			
	Cont	0.75±.03	0.63±.03	0.58 <u>+</u> .04	0.54±.03	0.62 <u>+</u> 03			
High	Adj	0.69±.06	0.59 <u>+</u> .04	0.51 <u>+</u> .03	0.56 <u>+</u> .02	0.58 <u>+</u> 03			
	Tg	0.74 <u>+</u> .04	0.69 <u>+</u> .04	0.61 <u>+</u> .04	0.61±.01	0.63±03			

Table (19) Analysis of variance for Data represented in table (18)

Source of variation	D.F	Sum of squares	Mean squares	F
Sex (S)	1	.082	.082	5.567**
Protein level (p)	2	.060	.030	2.051
Immunization (M)	2	.024	.012	.825
Interval (I)	4	4.805	1.201	81.363***
SxP	2	.035	.017	1.217
S×M	2	.077	.038	2.623
SxI	4	.071	.017	1.204
P x M	4	.072	.018	1.224
PxI	8	.296	.037	2.506**
MxI	8	.151	.018	1.287
Remainder	637	9.404	.014	



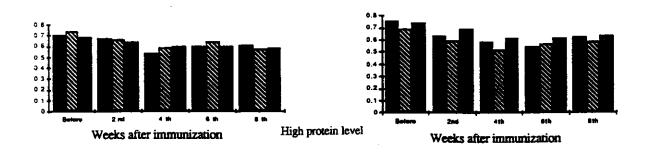
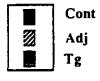


Fig (22) Albumin / globulin ratio (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.



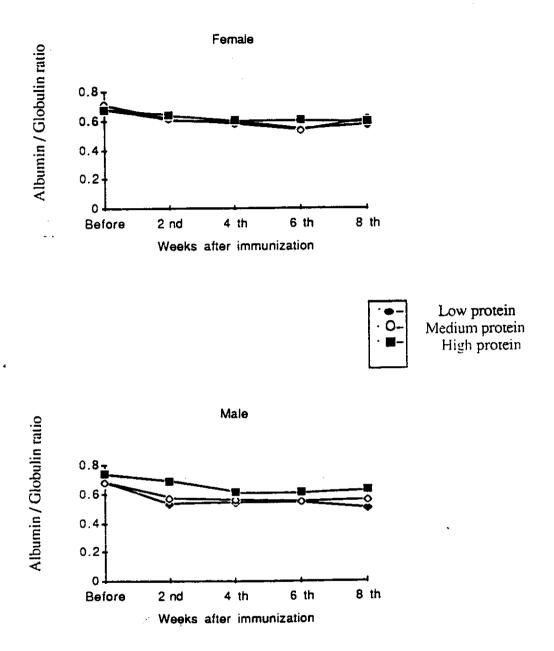


Fig (23) Albumin / globulin ratio (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

4.2.5 Serum total lipids

Data concerning the effect of dietary protein and induced thyroid immunization, chick's sex and time intervals on serum total lipids and presented in table (20) and illustrated in figures 24 and 25.

Obtained results indicated that serum total lipids were significantly affected by levels of protein in diet and time intervals. However, no signifecant effects were observed due to either chick's sex or thyroglobulin immunization (ANOVA table 21).

In most experimental groups, chicks fed higher levels of protein in diet had the lowest mean values of serum total lipids, while those fed low levels had the highest levels of serum lipids. Chicks recieved medium level of protein in diet were intermediate, this trend was observed in both female and male chicks.

Serum total lipids grand mean values were 472.6, 464.8 and 445.4mg/dl for female chicks fed low, medium and high protein levels, respectively. The corresponding values in males were 462.7, 450.0 and 450.0 mg / dl.

Slight increase in serum total lipids mean values was observed with the progress of time. Mean value for females averaged 417.3, 449.8, 463.7, 447.0 and 501, 8 mg/dl before and at $2^{\rm nd}$ $4^{\rm th}$, $6^{\rm th}$ and $8^{\rm th}$ weeks after immunization, the corresponding values in males were 440.7, 447.9, 451.3, 480.7 and 518.8 mg/dl, respectively.

Significant differences (P < 0.05) were noted due to the interactions between sex x dietary protein levels and immunity treatment x time intervals

4.2.6 Serum total cholesterol

Data Concerning the effect of protein levels in diet and thyroglobulin immunization on serum cholesterol for females and males Hubbard chicks at various time intervals was presented in table (22) and illustrated in figures 26 and 27.

Significant difference due to the time intervals was observed in serum level of cholesterol. On the other hand, no significant effect due to any of the studied factors was noted (ANOVA table 23).

Slight increase in serum cholesterol mean values were noted the group fed low protein diet than the higher ones, Decreased levels were observed in female immunized group in the last two intervals than the control or adjuvant injected chicks (Table 17).

With the progress of time decreased levels in serum cholesterol grand means were noted with different rates in females than in males, It averaged 148.8, 142.3, 133.9, 132.6 and 123.9 mg/dl from before to 2nd, 4th,6th and 8th weeks after immunization for female chicks, while the corresponding values were 144.7, 140.8, 118,1, 133.4 and 124.3 mg/dl for males, respectively,

Signifecant interaction effect between sex and intervals was obtained (ANOVA table 23) indicating that intervals affect serum cholesterol with different manner in females than males.

Results of the present study indicate that protein nutritional status influences serum total lipids, chicks fed low or deficient protein diet had increased levels of serum total lipids, while low concentrations of lipids in serum was always associated with feeding chicks high levels of protein in diets

Similar results were reported in rats (Mayer et al., 1956 and Ezzat et al., 1989) in rabbits, (Golden et al., 1954) in and chicken (Radbard et al., 1951).

The mechainsms by which low / or deficient protein diet increase plasma lipids is unclear, but it may be attributed to the continuous change in lipoprotein composition, transport and / or metabolism. On the other hand, it may be explained by the "compansatory effect" that might be done by birds to consume more feed to compansate the protein deficiency in diet which in turn

accumulate in serum as lipoproteins or total lipids and finally deposited as stored body fats.

These results were in agreement with Bryan and Jensen, (1989) who found that chicks fed 22% of crude protein in diet deposited significantly less fat than those fed lower levels of crude protein in diet.

No remarkable change was noted in serum cholesterol in the group of chicks fed either medium or high level of protein in diet. Chicks fed lower level of protein showed slightly higher cholesterol concentration in serum. Similar results were obtained by Radbard et al.,(1951) who indicated that protein malnutrition in chickens is accumpained by cholesterolenemia, It has been also observed in man (Beneridage et al., 1963). This may be attributed also to the increased mobilization of stored fat.

The significant effect due to time intervals which had been observed in females than males in serum cholesterol may be explained by the increasing demand of cholesterol for steriod hormones biosynthesis with the progress of age in unmatured female chicks.

The insignifecant effects obtained in serum choleserol and total lipids due to immunization may be explained by the unclear role of both of them in the sequence of events of the immune system, This was in agreement with Ibrahim et al., (1988).

Table (20)Effect of dietary protein levels on serum total lipids mg/dl $(X\pm S.E)$ of Female and Male chicks at various time intervals before and after thyroglobulin immunization.

TOTAL LIPIDS, mg / dl									
Protein	Group	Before	Weeks after immunization						
level	Group		2 nd	4 th	6 th	8 th			
FEMALE									
Low	Cont	459.5 <u>+</u> 18	461.0 <u>+</u> 28	483.9 <u>+</u> 19	491.5 <u>+</u> 34	475.8 <u>+</u> 29			
	Adj	423.2 <u>+</u> 19	456.7 <u>+</u> 32	480.5 <u>+</u> 17	496.1 <u>+</u> 19	476.8 <u>±</u> 14			
	Tg	453.5 <u>+</u> 16	476.3 <u>+</u> 28	494.5 <u>+</u> 22	488.7 <u>+</u> 32	526.4 <u>+</u> 23			
Medium	Cont	421.2 <u>+</u> 16	438.5 <u>+</u> 14	448.6 <u>+</u> 18	492.6 <u>±</u> 15	519.4 <u>+</u> 30			
	Adj	423.5 <u>+</u> 23	428.3 <u>+</u> 28	461.6 <u>+</u> 24	455.9 <u>+</u> 13	518.4 <u>+</u> 35			
	Tg	431.5 <u>±</u> 11	464.3 <u>+</u> 25	474.3 <u>+</u> 27	476.1 <u>+</u> 20	519.3±18			
High	Cont	409.9 <u>+</u> 29		İ	i	505.0 <u>+</u> 16			
	Adj	355.4 <u>+</u> 17	_	448.3 <u>+</u> 26	_	_			
	Tg	386.1 <u>+</u> 29	<u> </u>	421.9 <u>+</u> 22	485.0 <u>+</u> 21	568.7 <u>+</u> 16			
MALE									
Low	Cont	492.5 <u>+</u> 12	-	_		525.3 <u>+</u> 35			
	Adj	462.0 <u>+</u> 18	472.7 <u>+</u> 16	421.7 <u>+</u> 20	495.3 <u>±</u> 18	566.5 <u>+</u> 28			
	Tg	458.6 <u>+</u> 25	482.9 <u>+</u> 21	443.1 <u>±</u> 16	493.5 <u>+</u> 23	559.6 <u>+</u> 38			
Medium	Cont	469.0 <u>+</u> 16	475.4 <u>±</u> 18	469.4 <u>+</u> 18	455.4 <u>+</u> 24	485.1 <u>±</u> 31			
	Adj	470.3 <u>+</u> 9	462.5 <u>+</u> .14	489.3 <u>+</u> 15	401.0 <u>+</u> 29	534.6 <u>+</u> 32			
	Tg	443.6 <u>+</u> 17	466.0 <u>±</u> 12	434.7 <u>±</u> 15	548.8±10	501.4 <u>+</u> 24			
High	Cont	384.1 <u>+</u> 29	397.1 <u>+</u> 10	437.8 <u>+</u> 19	483.6 <u>+</u> 15	515.8 <u>+</u> 21			
	Adj	323.1 <u>±</u> 10	346.2 <u>+</u> 14	455.5 <u>+</u> 19	470.6 <u>+</u> 18	404.7 <u>+</u> 26			
	Tg	463.5 <u>+</u> 29	489.2 <u>+</u> 18	457.8 <u>+</u> 24	458.8 <u>+</u> 10	576.7 <u>+</u> 23			

Table (21) Analysis of variance for the Data represented in table (20)

Source of variation	D.F	Sum of squares	Mean squares	F
Sex (S)	1	3935.786	3935.762	.990
Protein level (p)	2.	125329.904	62664.95	14.808***
Immunization (M)	2	7961.635	3980.81	.941
Interval (I)	4	675610.544	168902.63	39.912***
SxP	2	27136.526	13568.263	3.216*
SxM	2	13683.143	6841.571	1.617
SxI	4	22798.130	5699.532	1.317
PxM	4	11905.494	2976.373	1.703
PxI	8	62238.115	7779.76	1.833
MxI	8	64814.949	8101.868	1.914*
Remainder	637	2695688.173	4231.84	

^{*} P < 0.05

^{**} P < 0.01

^{***} P < 0.001

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Male

Weeks after immunization

Female

Weeks after immunization

Fig (24) Serum Total lipids (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.

High protein level



Serum Total lipids mg / db

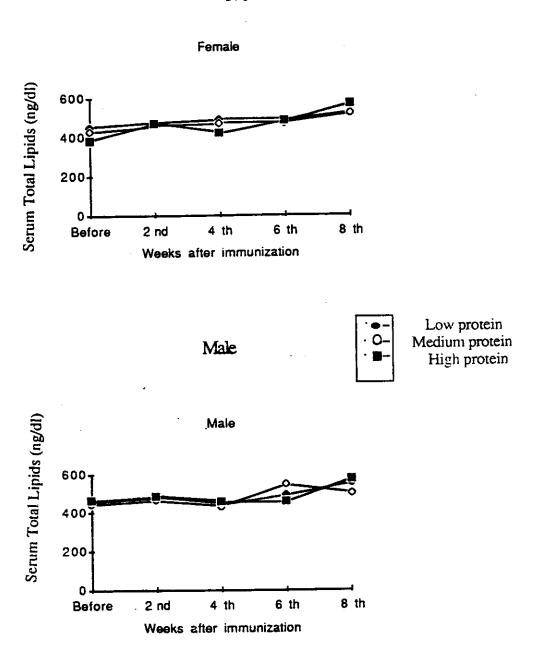


Fig (25) Serum Total Lipids (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

Table (22) Effect of dietary Protein levelson serum total cholesterol mg/dl $(\bar{X} \pm S.E)$ for Female and Male treated chicks at various time intervals before and after thyroglobulin immunization.

	SERUM TOTAL CHOLESTEROL mg/dl							
Protein		Defens	Weeks a	fter immi				
level	Group	Before	2 nd	4 th	6 th	8 th		
FEMALE								
	Cont	152.1 <u>+</u> 5.6	126.4 <u>+</u> 5.4	133.4 <u>+</u> 4.9	122.4 <u>+</u> 3.7	123.0 <u>+</u> 3.9		
Low	Adj	150.6 <u>+</u> 4.5	137.5 <u>+</u> 5.8	134.0±3.9	133.7 <u>+</u> 6.7	115.7 <u>+</u> 2.8		
	Tg	154.4 <u>+</u> 3.9	133.7 <u>+</u> 5.7	135.1±2.7	132.2 <u>+</u> 5.1	126.3 <u>+</u> 2.5		
	Cont	142.7±3.1	139.9 <u>+</u> 6.9	123.6 <u>+</u> 5.5	135.9 <u>+</u> 2.8	126.7 <u>+</u> 5.2		
Medium	Adj	148.7 <u>+</u> 3.1	130.4 <u>+</u> 5.4	138.1 <u>+</u> 5.6	131.6 <u>+</u> 4.7	123.5±4.8		
	Tg	148.4 <u>+</u> 2.9	130.7 <u>+</u> 6.7	133.7 <u>+</u> 4.5	133.4 <u>+</u> 6.2	125.5 <u>+</u> 5.4		
	Cont	146.0 <u>+</u> 3.7	}	1	133.0±3.7	1		
High	Adj	146.1 <u>+</u> 3.8			138.6±4.5			
	Tg	150.3 <u>+</u> 3.4	124.9 <u>+</u> 6.9	131.8 <u>+</u> 8.9	132.2 <u>+</u> 4.2	119.9 <u>+</u> 6.1		
			MALE		T			
	Cont	152.6 <u>+</u> 2.8	143.3+5.3	123.6 <u>+</u> 4.6	139.8 <u>+</u> 5.1	119.2 <u>+</u> 5.1		
Low	Adj	146.2 <u>+</u> 5.8	138.6 <u>+</u> 3.4	135.5 <u>+</u> 5.4	137.6±5.2	131.4 <u>+</u> 3.8		
	Tg	156.4 <u>+</u> 5.6	148.4 <u>+</u> 3.8	132.1 <u>+</u> 4.2	131.8±4.1	123.0 <u>+</u> 4.6		
	Cont	149.9 <u>+</u> 3.6	144.3 <u>+</u> 4.8	133.2 <u>+</u> 3.5	146.7 <u>+</u> 5.1	114.4 <u>+</u> 2.8		
Medium	Adj	147.6 <u>+</u> 4.6	138.2 <u>+</u> 3.4	135.4 <u>±</u> 4.5	140.8 <u>+</u> 5.7	123.8 <u>+</u> 2.6		
	Tg	144.6 <u>+</u> 5.1	140.5 <u>+</u> 4.4	137.4 <u>+</u> 5.7	132.9±3.2	124.2 <u>+</u> 4.3		
	Cont	142.7 <u>+</u> 2.3	135.5 <u>+</u> 7.2	129.1±3.4	126.3 <u>±</u> 4.1	124.6±6.1		
High	Adj -	145.6 <u>+</u> 4.6	140.7 <u>±</u> 5.6	130.9 <u>±</u> 7.2	125.2 <u>+</u> 4.5	130.6 <u>+</u> 4.0		
	Tg	144.1 <u>+</u> 3.3	139.1 <u>+</u> 8.9	136.5 <u>±</u> 3.4	126.2 <u>±</u> 4.7	126.6±4.8		

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Table (23) Analysis of variance for the Data represented in table (22)

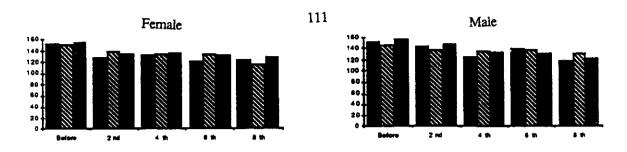
			M	1
Source of variation	D.F	Sum of squares	Mean squares	F
Sex (S)	1	368.369	368.369	2.084
Protein level (p)	2	110.069	55.034	.311
Immunization (M)	2	345.146	172.573	.967
Interval (I)	4	45491.942	11372.985	64.331***
SxP	2	486.740	243.370	1.377
SxM	2	14.006	7.003	.040
SxI	4	2822.971	705.742	3.992**
PxM	4	389.421	97.355	.551
PxI	8	2574.89	321.861	1.821
MxI	8	629.704	78.713	.445
Remainder	637	112614.071	176.788	

^{*} P < 0.05

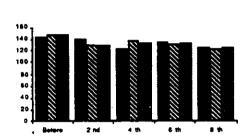
^{**} P < 0.01

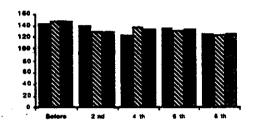
^{***} P < 0.001



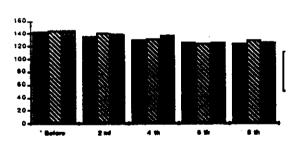


Low protein level





Medium protein level





Weeks after immunization

High protein level

Weeks after immunization

Fig (26)Serum Total Cholesterol (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.

Cont
Adj
Tg

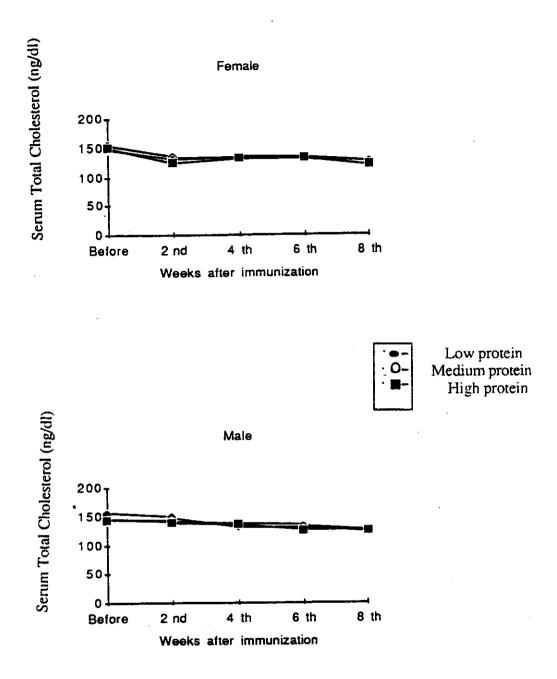


Fig (27) Serum Total Cholesterol (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

4.2.7 Serum Tranaminase activity (SGOT)

Data concerning the effect of dietary protein level and immunization for female and male chicks at various time intervals on serum glutamic - oxoloacetic transaminase activity (SGOT) are presented in table (24) and illustrated in figures 28 and 29.

Obtained results indicated that SGOT was significantly affected with chick's sex, dietary protein level, thyroglobulin immunization and time intervals (ANOVA table 25).

Regardless of any other factors, female chicks had higher values of SGOT than males, It averaged 30.97 and 28.5 u/ml for female and male chicks, respectively.

Protein level in chick's diet had pronouned effect on SGOT levels. Female chicks fed relatively low level of protein in diet had higher levels of the enzyme activities. It's grand means values were (32.2, 30.7 and 29.3 u/ml) for female chicks fed low, medium and high levels of protein in diet while the corresponding values of males were (30.9, 29.4 and 28.1 u/ml), respectively.

Variations in SGOT mean values due immunization were found to be indicative, control and adjuvant injected birds had approximate similar. grand mean values which were (30.5, 29.3 and 33.4 u/ml) for control, adjuvant and immunized females, While the corresponding values in males were (29.3, 29.0 and 31.4 u/ml), respectively.

Slight variations were noted with progress of time, however it were significant. Female grand means were 29.1, 29.3, 31.2 31.4 and 31.8 u/ml before and at the $2^{\rm nd}$, $4^{\rm th}$, $6^{\rm th}$, and $8^{\rm th}$, week after immunization .The corresponding values in males were 20.0, 23.4, 29.7, 29.9 and 31.2 u/ml respectively .

Significant interaction effects were observed between sex x interval and protein level x intervals (ANOVA table 25).

4.2.8. Serum Transaminase activity (SGpt)

The effect of dietary protein protein level and thyroglobulin immunization

on serum glutamic - pyruvic transaminase activity (SGpT) for both female and male Hubbard chicks are presented in table (26) and illustrated in figures 30 and 31.

Analysis of variance revealed significant effect due to protein level, time intervals and immunization on SGpT mean values (ANOVA table 27), However, no significant effect due to sex was observed.

Serum GPT grand mean values were of females 22.1, 20.9 and 21.1 u/ml for chicks fed low, medium and high level of protein, respectively. The corresponding values in males were 21.2, 20.4 and 20.5 u/ml, respectively.

Time intervals were observed to affect GPT activities, which were 21.1, 20.9, 21.9, 21.6 and 21.4 u/ml from before and at 2nd, 4th, 6th and 8th week after immunization, while the corresponding values in males were 21.2, 21.3, 21.4, 21.5 and 20.9 u/ml, respectively.

Significant effect was found due to the interaction between sex and time intervals indicated, that serum level of GpT affected by time in female chicks in different manner than males.

Under the present exprimental conditions estimations of serum levels of transaminas enzymes (GOT and GPT) demonstrated that the level of protein in diet influenced both of them.

Chicks fed low or deficient level of protein in diet showed higher levels of GOT, GPT which are generally indicative to amino acids transformation, Bell and Sturkie, (1965) stated that the first stage in catabolism of amino acid is the formation of the corresponding keto acid and the nitrogen under the influence of transaminases activities in case of severe protein deficiency. This was agreement with the results of (Heibashy, 1990) in rats. While ,Gessler, (1965) found that increasing dietary protein level did not influence the activity of SGpt in plasma.

The higher values in SGoT, SGpT exisit in thyrogloblin immunized groups were in agreement with Odette, (1986) and Ibrahim et al., (1988), It may be attributed to the continuous need for amino acids transamination to compansate the protein deficiency in diet.

Table (24)Effect of dietary protein levelson serum Transaminase activity SGOT μ/ml ($\overline{X}\pm S.E$) of Female and Male chicks at various time intervals before and after thyroglobulim immunization .

SER	UM GLI	JTAMIC C	XALO AC	CETIC AC	ID ACTI	VITY	
Protein	Group	Before	Weeks after immunization				
level	Group	Berore	2 nd	4 th	6 th	8 th	
<i>y</i>			FEMALE			· 	
	Cont	27.6±1.2	30.8 <u>+</u> 1.0	33.9 <u>+</u> 1.2	31.2±1.6	32.5 <u>+</u> 1.9	
Low	Adj	28.7 <u>+</u> 1.2	30.2±1.3	31.8±2.1	33.4 <u>+</u> 2.1	36.3±2.4	
	Tg	30.1±.96	31.5 <u>+</u> .84	34.6 <u>+</u> 2.8	35.3±1.1	35.6 <u>+</u> 1.0	
	Cont	30.2±1.1	29.9 <u>+</u> .44	30.1 <u>+</u> .93	29.5±1.3	31.1 <u>+</u> .89	
Medium	Adj	31.3±1.0	26.9 <u>+</u> .35	33.1 <u>+</u> 1.4	30.7 <u>+</u> 1.4	30.3±1.2	
	Tg	29.5 <u>+</u> 1.2	29.1±1.4	33.1±.73	32.1±1.4	34.4 <u>+</u> 1.3	
•	Cont	28.6±1.6	29.8±.66	29.7±.49	31.7 <u>±</u> .80	31.7 <u>±</u> 1.2	
High	Adj	27.7 <u>+</u> 1.5	26.8 <u>+</u> .67	28.6±1.3	30.1±.78	30.3±1.2	
1	Tg	28.4±1.3	29.6±.72	26.2 <u>+</u> .56	29.6±.57	30.5±.94	
			MALE				
	Cont	28.7±1.2	31.5 <u>±</u> 1.8	30.6 <u>+</u> 2.1	30.3 <u>+</u> 2.2	30.6 <u>+</u> .84	
Low	Adj	29.9 <u>+</u> 1.1	29.9 <u>+</u> 1.9	31.3 <u>+</u> 1.5	27.6 <u>+</u> 2.4	34.5 <u>+</u> 1.4	
	Tg	31.3±1.5	29.8±1.3	32.0 <u>+</u> 1.1	31.4 <u>+</u> 2.2	34.6 <u>+</u> .85	
	Cont	29.7±1.1	28.4 <u>+</u> .53	28.2 <u>+</u> 1.3	27.6 <u>+</u> 1.2	30.7±1.9	
Medium	Adj	29.7±1.5	29.7±1.4	26.7±1.3	29.8±1.5	32.2±2.7	
	Tg	29.6±1.3	28.6±.71	29.0±.43	31.6±.14	29.7±1.1	
	Cont	26.7±1.5	27.3±.88	28.7 <u>+</u> 1.2	.30.9±.80	30.1±.84	
High	Adj	28.3±1.6	29.2 <u>+</u> 1.1	27.8±.40	28.8±.37	28.2±1.3	
	Tg	28.1 <u>+</u> 2.4	30.6 <u>+</u> .44	28.9 <u>+</u> .60	31.1±1.7	30.0±.55	

Table (25) Analysis of variance for the Data represented in table (24)

Source of variation	D.F	Sum of squares	Mean squares	F
Sex (S)	1	335.723	335.723	23.878***
Protein level (p)	2	645.538	322.769	22.956***
Immunization (M)	2	161.616	80.808	5.747**
Interval (I)	4	1150.822	287.705	20.463***
SxP	2	2.362	1.181	.084
SxM	2	3.823	1.91	.136
SxI	4	140.855	35.213	2.505*
PxM	4	23.346	5.836	.415
PxI	8	431.535	53.941	3.837***
MxI	8	176.953	22.106	1.572
Remainder	637	8956.270	14.060	

^{*} P < 0.05

^{**} P < 0.01

^{***} P < 0.001

Low protein level

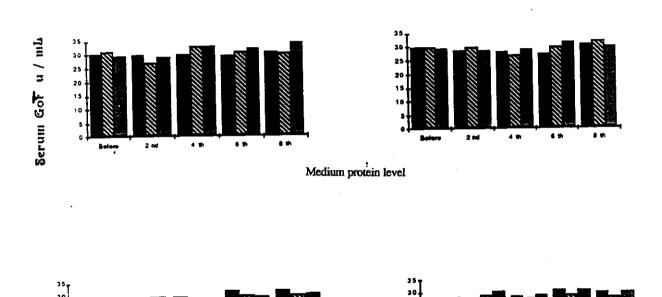


Fig (28) Serum Got (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.

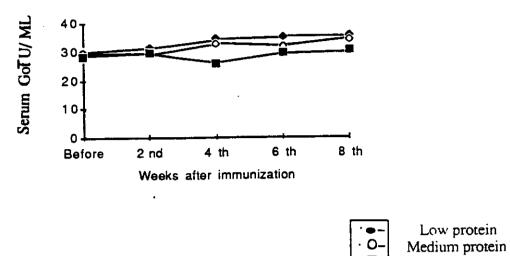
High protein level

Weeks after immunization

	Cont
Z	Adj
	Тg

Weeks after immunization







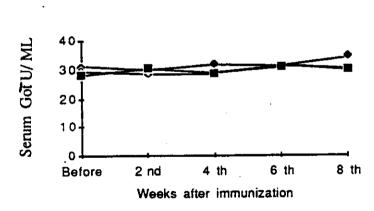


Fig (29) Serum GoT (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

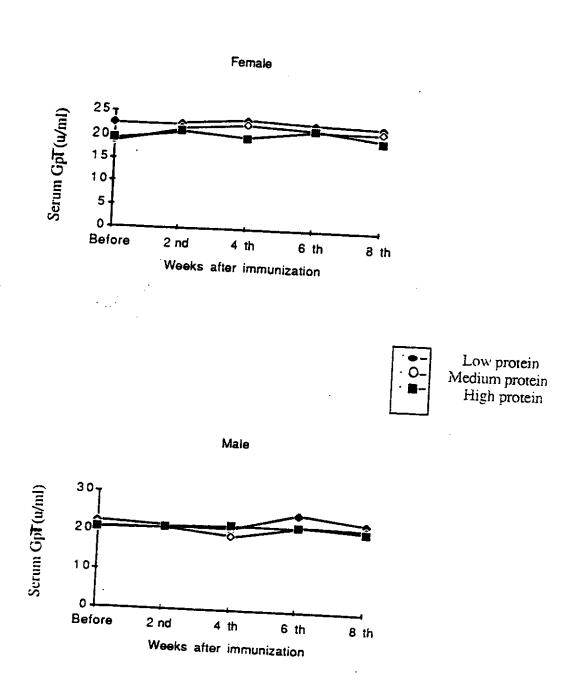


Fig (31) Serum GpT (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

3 - Effect of dietary protein level on some growth measurments

Body weight, the body gain, feed consumption and feed efficiency were considered to be as criteria for growth and performance for both female and male chicks at the 5^{th} , 8^{th} , 10^{th} , 12^{th} and 14^{th} week of age which were equal to before and at the 2^{nd} , 4^{th} , 6^{th} and 8^{th} week after immunization.

4.3.1 Body weight.

The average body weights in all groups studied are presented in table (28) and illustrated in figures 32 and 33.

Obtained results showed significant effects (P < 0.001) due to all studied factors (i.e. sex dietary protein levels, thyroglobulin immunization and time intervals) on chick's body weight.

A significant effect due to the interactions between sex and both dietary protein level and immunization and between Intervals and both dietary protein level and sex were observed (ANOVA table29)

Regardless of any other factor, female chicks always had lighter body weights than males. Females grand mean values were 1909.9 g and 2213.3 g for males.

Protein level in chick's diet significantly affect the body weight, chicks fed low level of protein in diet had the lightest weights, this was evident in both female and male chicks. Grand mean body weight amounted to 1618.6, 1916.7 and 2192.3 g for female chicks fed low, medium and high dietary protein level, respectively. The corresponding values were 1702.3, 2095,5 and 2337.7 g in males, respectively.

Thyroglobulin immunized group of chicks showed slightly heavier average body weights than those of control or adjuvant injected group. Female chicks

averaged 1984.4, 1908.5 and 1874.3 g for throglobulin immunized, adjuvant and control group, respectively. The corresponding values of males were 2049.3, 1859.8 and 1933.8 g, respectively.

Increased body weights were noted with the progress of time, females had slower rate of growth than males. Average body weights of females were 879.5 ,1685.1 , 2007.4 , 2328.2 and 2626.3 g from before and at the $2^{\rm nd}$, $4^{\rm th}$, $6^{\rm th}$ and $8^{\rm th}$ week after immunization , respectively. The corresponding values in males were 891.8 , 1743.5 , 2105.7 , 2412.8 and 2739.5 gms ,respectively .

Under the present experimental condition, it was clearly observed that protein level in diet affect both female and male body weight, Chicks fed low level of protein in diet gained less body weights in comparison with those fed medium or higher protein level in diet, particularly up to the 8th week of age.

The increase in body weight were found to be associated with the increasing percent of protein in diet. In the present study, birds fed 18.41% crude protein in diet attained heavier weights than those fed 14.14%. While no gross difference in chicks weight were observed between the former and those birds fed 22.11% crude protein in diets during the period from 21 - 56 days.

Several reports have indicated that optimal broiler growth can be obtained with crude protein level in diet lower than 18% (Hurwitz et al., 1980; Edmond et al., (1985); Nakajma et al., (1985) and Uze (1986). However, Wilson et al., (1987) demonstrated that level of 12% crude protein could be fed to broiler males without adversely affecting body weights.

Similar results were reported by Payne et al., (1990) and Mario and Waldrop, (1991) who found that body weight of both female and male chicks were significantly influenced by dietary protein restriction, similar observations were also found by Parr and Summers, (1991).

A retarded growth rate as a result of feeding lower levels of crude protein may be attributed to either insufficient or imbalanced amino acids contents in diet. Thismay affect the protein synthesis and metabolic rate during the early phase of growth.

The present study also revealed that no great difference was observed in body weight of chicks fed 14% and those 18% crude protein in diet. This may be explained on the light of the compansatory response to early protein deficiences. Moran, (1973) and Pesti and Fletcher, (1984) suggested that chickens have the ability to correct earlier protein deficiences through a phenomenon called compansatory growth.

Continuous feeding chicks higher level of crude protein significantly improved body weight gains, which was clearly observed at 8th week of age (market age), however, this concomitant increase the total meat yield which indicated that body weight gains was directly related to dietary protein (Stilborn and Waldrop, 1988). In agreement with the previouse results were reported by Yoshida and Morimato, (1970) who added that broilers fed higher crude protein levels deposited significantly less abdominal fat and high net meat yield than those fed lower crude protein which deposited significantly more fats.

It could be concluded that the decrease in body weight due to low dietary proteindoes not necessarily imply that a level greater than 18% of protein in diet is needed to maintain acceptable performance. However, it implies the need to maintain the minimum amino acid content in which are essential different metabolic processes.

Feeding higher levels of protein in diet was associated with the increased production of antibodies against thyroglobulin immunization. It could be concluded that increasing the level of protein in diet does not imply increased body gain only but also means stronger response against invading antigens.

Table (28)Effect of dietary protein levels on Body weight /g. $(X \pm SE)$ of female and male treated chicks at various time intervals before and after thyroglobulin immunization

	BODY WEIGHT (g)								
Protein	Group	Before	Weeks after immunization						
level	Group	Delore	2 nd	4 th	6 th	8 th			
	FEMALE								
	Cont	652 ± 19	1471 <u>+</u> 20	1773 <u>+</u> 30	2066±20	2283±40			
Low	Adj	714 <u>+</u> 29	1391 <u>+</u> 44	1643 <u>±</u> 44	1973 <u>+</u> 43	2271±43			
	Tg	650 <u>+</u> 19	1419 <u>+</u> 30	1641 <u>+</u> 38	2014 <u>+</u> 31	2318 <u>+</u> 41			
	Cont	823 <u>+</u> 23	1631 <u>+</u> 27	1971 <u>+</u> 24	2274 <u>+</u> 21	2504 <u>±</u> 30			
Medium	Adj	955 <u>+</u> 45	1683 <u>+</u> 41	2000 <u>+</u> 34	2300 <u>+</u> 57	2614 <u>+</u> 39			
	Tg	966 <u>+</u> 25	1751 <u>+</u> 32	2091 <u>+</u> 28	2321 <u>+</u> 28	2668 <u>+</u> 46			
	Cont	1101 <u>+</u> 45	1850 <u>+</u> 24	2188 <u>+</u> 23	2587 <u>+</u> 27	2936 <u>+</u> 76			
High	Adj	949 <u>+</u> 30	2014 <u>+</u> 41	2380 <u>+</u> 28	2713 <u>+</u> 30	3028 <u>+</u> 36			
	Tg	1088 <u>+</u> 22	1956 <u>+</u> 31	2387 <u>+</u> 29	2706 <u>+</u> 28	3014 <u>+</u> 44			
	· <u>·</u>	<u>.</u>	MALE						
	Cont	691 <u>+</u> 47	1485 <u>+</u> 37	: 1867 <u>+</u> 33	2177 <u>±</u> 35	2437 <u>+</u> 42			
Low	Adj	783 <u>+</u> 27	1489 <u>+</u> 66	1772 <u>+</u> 17	1981 <u>+</u> 32	2352 <u>+</u> 53			
	Тд	701 <u>+</u> 39	1563 <u>+</u> 53	! 1894 <u>+</u> 43	2133±29	2510 <u>+</u> 33			
	Cont	780 <u>+</u> 52	1611 <u>+</u> 32	1998 <u>+</u> 55	2298 <u>±</u> 51	2661 <u>+</u> 45			
Medium	Adj	792 <u>+</u> 34	1607 <u>+</u> 24	1967 <u>+</u> 23	2288 <u>+</u> 34	2653 <u>+</u> 23			
	Tg	947 <u>±</u> 16	1802 <u>±</u> 26	2154 <u>+</u> 15	2457 <u>+</u> 25	2723 <u>+</u> 28			
	Cont	1078 <u>+</u> 12	1974 <u>±</u> 10	2286 <u>+</u> 24	2689 <u>+</u> 23	2976 <u>+</u> 49			
High	Adj	1090 <u>+</u> 29	2032±23	2463 <u>+</u> 61	2850 <u>+</u> 65	3177 <u>+</u> 26			
	Tg	1165±17	2123 <u>+</u> 29	2551 <u>+</u> 24	2843 <u>+</u> 24	3164 <u>+</u> 83			

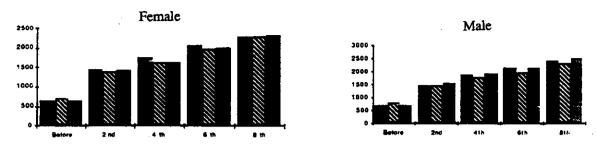
Table (29) Analysis of variance for Data represented in table (28)

Source of variation	D.F	Sum of squares	Mean square	F
Sex (S)	1	2301831.40	2301831.40	185.90***
protein level (p)	2	87928254.83	43964127.41	3550.76***
Immunization (M)	2	278634.04	139317.02	11.25***
Intervals (I)	4	541381389.07	135345347.26	10931.18***
SxP	2	501359.03	250697.51	20.24***
SxM	2	104503.64	52251.82	4.22**
SxI	4	245064.55	61266.13	4.94***
PxM	4	76276.32	19069.08	1.540
РхI	8	11233842.06	1404230.25	113.41***
M'x I	8	86400.46	10800.05	.872
Remainder	637	7887066.91	12381.58	

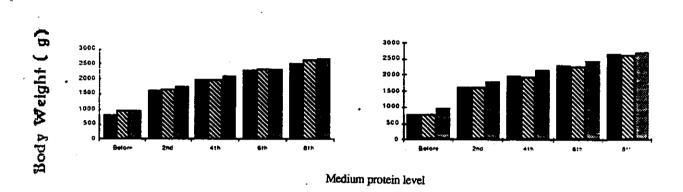
^{*} P < 0.05

^{**} P < 0.01

^{***} P < 0.001



Low protein level



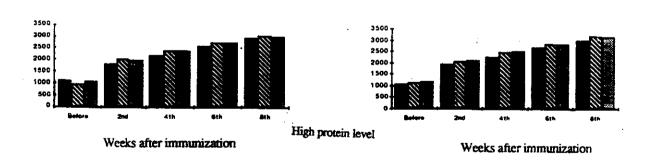


Fig (32) Body Weight (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.



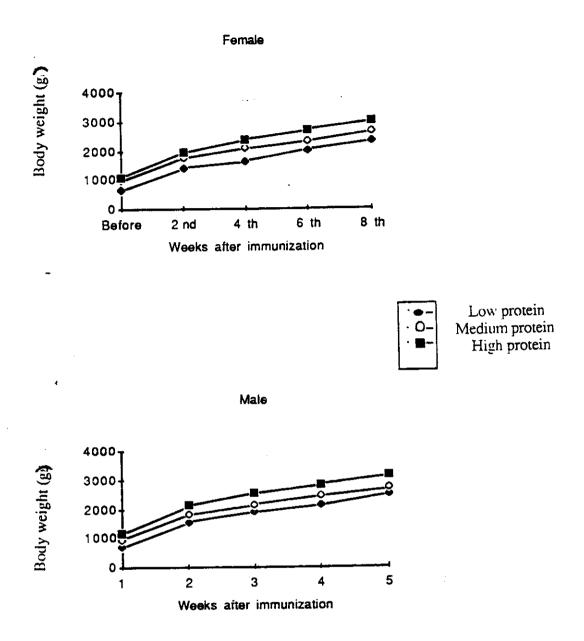


Fig (33) Body Weight (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

4.3.2 Feed Consumption

The effect of protein level in diet and thyroglobulin immunization on feed consumption of both female and male chicks at various time intervals are presented in table (30) and illustrated in figures 34 and 35.

Feed consumption was significantly affected by dietary protein and time intervals, No significant effects were noted due to either sex or active thyroid immunization (ANOVA table 31).

Total amount of feed consumed throughout the experimental period (14 weeks) amounted about 8662.3, 9126.5 and 9632.8 g for female chicks fed low, medium and high level of protein, respectively. The corresponding values for males were 8689.3, 9262.6 and 9767.3 gms, respectively.

Increasing values of feed consumption for both female and male chicks with the progress of time were observed. Average mean values of female feed consumption were 2061.1, 2438.3, 1517.2, 1559.3 and 1606.5 g before and at the $2^{\rm nd}$, $4^{\rm th}$, $6^{\rm th}$, and $8^{\rm th}$ week after immunization, respectively. (which were equal to the $5^{\rm th}$, $8^{\rm th}$, $10^{\rm th}$, $12^{\rm th}$ and $14^{\rm th}$ weeks of age)The corresponding values in males were 2065.1, 2489.3, 1527.6, 1586.3 and 1612.2 gms, respectively.

Significant interaction effects between time intervals and both protein levels in diet and immunization were observed and also between sex and dietary protein levels (ANOVA table 31)

The present results showed that birds fed low dietary protein level consumed more feed than those fed medium or higher levels. This may be due to the compansatory response, where the chicks consume more feed to compansate the protein deficiency. Waldroup et al., (1976) reported that growth potential of a broilers is largely governed by volentaly feed intake which in turn can be reduced if there is a suitable built up of plasma amino acid. Parr and Summers, (1991) were in agreement with the mentioned result,

Table (30) Effect of dietary protein levels on feed consumption/g/birdy

Week (X+S.E) of female and Male chicks at various time intervals

before and after thyroglobulin immunization.

	Feed Consumption, (g)								
Protein	Group	Before	Weeks at	ter immu	nization				
level			2 nd	4 th	6 th	8 th			
FEMALE									
<u> </u>	Cont	1849 <u>+</u> 33	2478 <u>+</u> 22	1408 <u>+</u> 5	1435 <u>+</u> 18	1400 <u>+</u> 17			
Low	Adj	1837 <u>+</u> 26	2412 <u>+</u> 20	1422 <u>+</u> 27	1472 <u>+</u> 32	1482 <u>+</u> 31			
	Tg	1816 <u>+</u> 32	2573 <u>±</u> 20	1394 <u>+</u> 11	1453 <u>+</u> 10	1463 <u>+</u> 17			
	Cont	. 2057 <u>+</u> 34	2313 <u>+</u> 35	1536 <u>+</u> 3	1577 <u>+</u> 34	1639 <u>+</u> 53			
Medium	Adj	2214 <u>+</u> 71	2256 <u>+</u> 66	1496 <u>+</u> 43	1559 <u>±</u> 21	1642 <u>+</u> 32			
,	Tg	2026 <u>+</u> 38	2446 <u>+</u> 64	1598 <u>+</u> 42	1560 <u>+</u> 37	1603 <u>+</u> 36			
	Cont	2267 <u>+</u> 55	2363 <u>+</u> 31	1611 <u>+</u> 34	1698 <u>+</u> 42	1740 <u>+</u> 36			
High	Adj	2356 <u>+</u> 72	2533 <u>+</u> 81	1695 <u>+</u> 50	1638 <u>+</u> 13	1729 <u>+</u> 72			
	Tg	2130 <u>±</u> 67	2571 <u>+</u> 60	1595 <u>+</u> 42	1609 <u>÷</u> 41	1707 <u>+</u> 51			
			MALE						
	Cont	1868.4±13	2440 <u>+</u> 14	1402.3 <u>+</u> 27	1421 <u>+</u> 18	1439 <u>+</u> 12			
Low	Adj	1958.8 <u>+</u> 51	2459.2 <u>+</u> 43	1453.3 <u>+</u> 33	1472 <u>+</u> 31	1481 <u>+</u> 42			
	Tg	1876.0 <u>+</u> 22	2456.5 <u>+</u> 24	1507.7 <u>+</u> 24	1462 <u>+</u> 17	1480 <u>+</u> 32			
	Cont	1905 <u>±</u> 37	2463 <u>+</u> 41	1521 <u>+</u> 40	1762 <u>+</u> 70	1627 <u>+</u> 64			
Medium	Adj	2177 <u>+</u> 63	2284 <u>+</u> 56	1516 <u>+</u> 34	1661 <u>+</u> 22	1650 <u>+</u> 31			
	Tg	2057 <u>±</u> 28	2446 <u>+</u> 64	1498 <u>+</u> 42	1544 <u>+</u> 45	1673 <u>+</u> 71			
	Cont	2350 <u>+</u> 50	2424 <u>+</u> 42	1627 <u>±</u> 33	1633 <u>+</u> 42	1730±50			
High	Adj	2391 <u>+</u> 60	2458 <u>+</u> 38	1616 <u>+</u> 51	1689 <u>+</u> 70	1701 <u>±</u> 36			
	Tg	2208 <u>+</u> 72	2440 <u>+</u> 63	1624 <u>+</u> 59	1682 <u>+</u> 47	1729 <u>+</u> 43			

Table (31) Analysis of variance for Data represented in table (30)

Source of variation	D.F	Sum of squares	Mean square	F
Sex (S)	1	25413.63	25413.63	1.67
protein level (p)	2	5818197.68	2909098.84	192.24***
Immunization (M)	2	18022.73	9011.36	.596
Intervals (I)	4	83778400.47	20944600.11	1384.12***
SxP	2	111766.52	55883.26	3.69*
SxM	2	10461.52	5230.76	.346
SxI	4	84083.05	21020.76	1.389
PxM	4	126678.36	31669.59	2.093
PxI	8	2445030.89	305628.86	20.19***
MxI	8	804171.64	100521.45	6.643***
Remainder	637	9639096.21	15132.01	

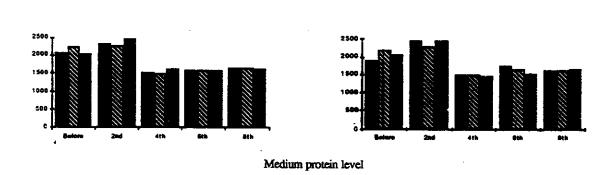
^{*} P < 0.05

^{**} P < 0.01

^{***} P < 0.001

Low protein level





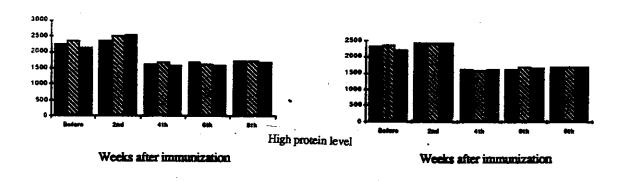


Fig (34) Feed Consumption (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.



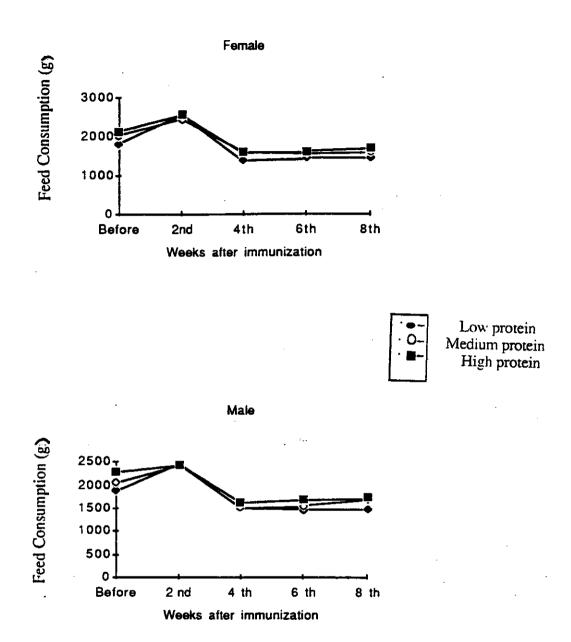


Fig (35) Feed Consumption (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals

4.3.3 Feed efficiency

Data concerning the effect of dietary protein level and induced thyroid immunization on feed efficiency for both female and male chicks at various time intervals were presented in table (32) and illustrated in figures 36 and 37.

Obtained results indicated that feed efficiency was significantly affected by bird's sex, protein level in diet and time intervals (ANOVA table 33)

Feed efficiency grand means amounted to 0.264, 0.288 and 0.300 for female chicks fed low, medium and high levels of protein, respectively. The corresponding values in males were 0.278, 0.296 and 0.327, respectively. However, males had always higher feed efficiency values than females.

Feed efficiency mean values were decreased sharply after the 8^{th} week of age up to the end of the experimental period (4^{th} , 6^{th} and 8^{th} weeks after immunization). This was quite true for both female and male chicks. As a function of intervals. The grand mean values of feed efficiency for females were (0.406, 0.381, 0.227, 0.199 and 0.204) from before and at the 2^{nd} , 4^{th} , 6^{th} and 8^{th} week after immunization, respectively. The corresponding values for males were 0.406, 0.381, 0.227, 0.199 and 0.204, respectively.

Significant interaction effects were observed due to sex and both dietary protein level and time intervals indicating that females had different response to either protein level or time intervals than males. A significant interaction effects have also been noted between dietary protein level and both immunization and time intervals.

Under the present experimental conditions feed efficiency was observed to be greatly affected by dietary protein level particularly up to the marketing age (8th week of age). While Chicks fed lower level of protein in diet (14.44%) attained lower values of feed efficiency, higher values were attained in the

group of chicks fed medium (18.41%) or high (22.11%) levels of protein in diet during the grower or finisher period.

Although body weight gain and feed efficiency seemed to be directly proportional to protein level in diet, benifits of continuous feeding on higher levels of protein seemed to have pronounced effect on body gain (e.g) improve feed efficiency. These results are in agreement with those of obtained by Jakson et al., $(1982)_{(a)}$ and Pasti and Fletcher, (1984) who observed improved feed efficiency with higher levels of protein in diet.

This may raise the question of the costs of feeding higher dietary protein level .Jakson et al., (1982)_(b) reported higher costs and lower net return when dietary protein levels were greater than (20%) had been used. Mario and Waldrop, (1991) were in disagreement with the last results they observed that the benefits of using continuous feeding on overall performance were limited to improve feed efficiency.

With increasing age, a remarkable decrease in feed efficiency was noted in all groups due to the higher amounts of feed consumed by birds or and lower body gain less requirements for amino acids in the older ages. were reported by Mario and Waldrop, (1991).

Table (32) Effect of dietary protein levels on Feed efficiency (g/g) mean values of Female and Male treated chicks at various time intervals before and after thyroglonbulin immunization.

FEED EFFICIENCY (g/g)								
Protein	_			ter immu	nization			
level	Group	Before	2 nd	4 th	6 th	8 th		
FEMALE								
	Cont	0.357	0.331	0.224	0.306	0.201		
Low	Adj	0.341	0.310	0.189	0.229	0.207		
	Tg	0.362	0.347	0.230	0.199	0.209		
	Cont	0.400	0.376	0.224	0.205	0.196		
Medium	Adj	0.431	0.381	0.187	0.213	0.198		
	Tg	0.443	0.391	0.231	0.201	0.217		
	Cont	0.432	0.413	0.221	0.234	0.215		
High	Adj	0.464	0.438	0.234	0.196	0.186		
	Tg	0.456	0.427	0.264	0.203	0.198		
			MALE					
	Cont	0.361	0.340	0.271	0.216	0.201		
Low	Adj	0.350	0.352	0.188	0.167	0.229		
	Tg	0.371	0.361	0.228	0.196	0.254		
	Cont	0.402	0.371	0.254	0.196	0.221		
Medium	Adj	0.389	0.356	0.231	0.193	0.218		
	Tg	0.421	0.403	0.239	0.201	0.168		
Hìgh	Cont	0.431	0.409	0.196	0.192	0.216		
	Adj	0.455	0.439	0.234	0.271	0.188		
	Tg	0.473	0.438	0.251	0.188	0.185		

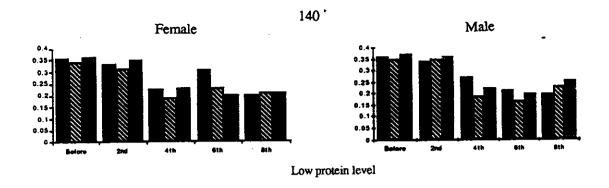
Table (33) Analysis of variance for Data represnted in table (32)

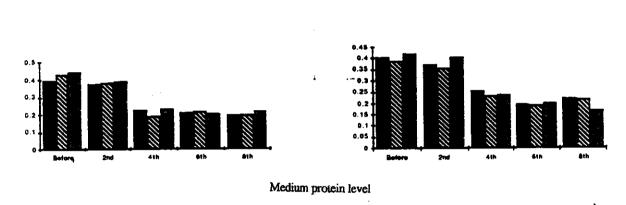
Source of variation	D.F	Sum of squares	Mean square	F
Sex (S)	1	36582.71	36582.71	56.60***
protein level (p)	2	793834.26	39 69 17.13	614.14***
Immunization (M)	2	3285.14	1642.57	2.54
Intervals (I)	4	1164002.18	291000.54	450.25***
SxP	2	10566.82	5283.41	8.175***
SxM	2	566.22	283.11	.438
SxI	4	6543.112	1635.77	2.531*
PxM	4	8173.94	2043.48	3.162**
, PxI	8	353256.50	44157.06	68.323***
МхI	8	4645.51	580.68	0.898
Remainder	637	411691.77	646.29	

^{*} P < 0.05

^{**} P < 0.01

^{***} P < 0.001





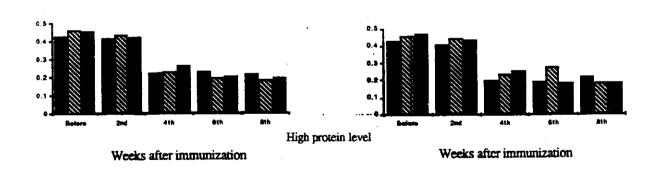


Fig (36) Feed efficiency (means) of Female and Male chicks at low, medium and high levels dietary protein before and after immunization.



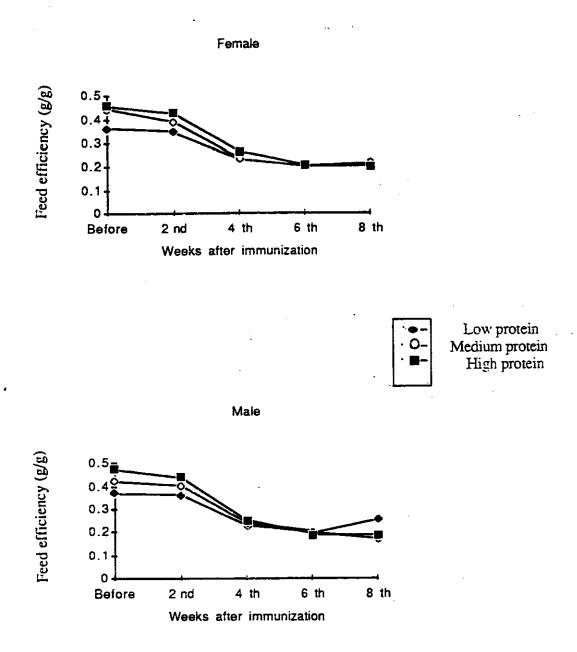


Fig (37) Feed efficiency (means) of thyroglobulin immunized Female and Male chicks fed low, medium and high dietary protein levels at various time intervals