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EFFECTS OF CA-AMINOPLEX INJECTION ON SOME PRODUCTIVE PERFORMANCE AND IMMUNE RESPONSE OF BROILER CHICKS

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ABSTRACT

One hundred twenty unsexed one-day old, Hubbard broiler chicks were randomly divided into 4 treatments in 3 replicates with 10 chicks each (4 treatments X 3 replicates X 10 chicks = 120 chicks). The experimental period was divided into two feeding phases, starter (from 0-3 weeks of age) and grower (from 4-6 weeks of age). The basal experimental diets were of 23.33 and 20.42% crude protein and 3115 and 3162 Kcal ME/kg diet for the starter and the grower diet, respectively. The experiment was included four treatments. Chicks in treatment 1 were not injected with Ca-Aminoplex and served as a control treatment; while chicks in treatment 2 were injected muscularly with 0.3 ml of Ca-Aminoplex at the beginning of the 2nd week, whereas chicks in treatment 3 were injected muscularly with 0.3 and 0.4 ml Ca-Aminoplex at the beginning of 2nd and 4th weeks, respectively. In treatment 4, chicks were injected muscularly with 0.3, 0.4 and 0.5 ml of Ca-Aminoplex at the beginning of 2nd, 4th and 6th weeks of age, respectively. At 6 weeks of age, five chicks from each treatment were injected intravenously with 0.2 ml of suspension of packed sheep red blood cells (SRBC). Sera were collected on the seventh day post immunization and antibody titer against SRBC was determined. Results obtained could be summarized as follow:

- All injected groups with Ca-Aminoplex were heavier (P<0.05) in body weight at the end of 2nd week than the control group, while at the end of 4th and 6th weeks the injected groups with Ca-Aminoplex two and three times being heavier (P<0.05) than the control and one injected groups.
- The injected groups with Ca-Aminoplex two or three times were higher (P<0.05) in daily gain and feed intake and better (P<0.05) in feed conversion at 4-6 and 0-6 weeks of age compared to control and injected one time groups.
- The injection with Ca-Aminoplex two or three times decreased (P<0.05) the cost of diet/kg gain, increased the cost of diet/ bird and recorded the highest (P<0.05) net revenue compared to the control or one injected groups.
- The injection of Ca-Aminoplex led to significantly (P<0.05) effect on Ab. in the serum at the 7th week of age, being higher in the group injected with Ca-Aminoplex three times than the other groups. Furthermore, the injected group with Ca-Aminoplex two times was higher (P<0.05) than the control and one injected groups.

It could be concluded that, using Ca-Aminoplex two or three times improved broiler performance and immune response.

INTRODUCTION

The amino acids are often referred to as the "building stones" from which the proteins are made. Synthetic amino acids are being used in increasing quantities to supplement the natural proteins.

Vitamins are a group of organic compounds found in foods in small quantities and they are an essential part of a good nutrition program. Adequate intake levels of vitamins are necessary for normal body functions,

growth and reproduction. Vitamin deficiencies can lead to a number of diseases or syndromes. The B-complex vitamins are involved in many metabolic functions including energy metabolism. It may be beneficial in some circumstances, such as birds subjected to stress. For normal productive nutritionists usually add a vitamin premix to the diet to compensate for fluctuating levels found naturally in food. Vitamin B6, although not an antioxidant, plays an important role in antioxidant defense by virtue of its metabolic role in the formation of cysteine, which is the rate-limiting precursor in the formation of glutathione (Grimble, 1998).

Nutrients are known to influence the responses of poultry to a disease challenge. Normally, during such a challenge nutrients are shunted away from growth. For example, body proteins are broken down and amino acids are shunted away from growth and are used by specific cells to synthesize critical proteins which allow the bird to mount a successful immune response to a particular disease challenge (Butcher and Miles, 2002).

Ca-Aminoplex was generally used in both animal and poultry as a calcium solution associated with amino acids and vitamins. It is of great help in case of calcium deficiency, restores the metabolic balance and favors recovery of convalescent animals. It is highly recommended for treating rickets and malnutrition. This drug provides vigor and reconstitutes and supplies energy favoring a rapid recovery.

The present study aimed to evaluate the effects of Ca-Aminoplex injection on the productive performance and immune response of broiler chicks.

MATERIALS AND METHODS

This study was carried out at the Poultry Farm, Department of Animal and Poultry Production, Faculty of Agriculture, South Valley University, in Qena during the period from May 2005 up to July 2005. It was designed to evaluate the effects of Ca-Aminoplex injection on broiler performance and immune response.

Ca-Aminoplex was purchased from Laboratorios Tornel (Mexico city) and its composition are presented in Table (1).

1- Experimental design:

One hundred twenty unsexed one-day old, Hubbard broiler chicks were purchased from a commercial local source. Chicks were randomly divided into 4 treatments in 3 replicates with 10 chicks each (4 treatments X 3 replicates X 10 chicks = 120 chicks). Chicks in each replicate within each treatment had nearly similar initial live body weight. Chicks were reared in two-tier wire floor battery in a windowless house. The chicks of each replicate were allocated in a cage with slatted floor of iron. The diameters of the cage were 70X70X75 cm for length, width and height, respectively. All cages were kept inside one room.

Table 1. Composition of Ca-Aminoplex.

Each 100 ml contain:	Amount
Calcium Gluconate	24 g
Boric Acid	4.8 g
D-Glucose	6 g
Vitamin B1 (Thiamine)	20 mg
Vitamin B2 (Riboflavin)	8 mg
Vitamin B3 (Panthenol)	8 mg
Vitamin B5 (Nicotinamide)	100 mg
Vitamin B6 (Pyridoxine)	20 mg
Amino Acids	10 mg
DL-Methionine	0.001g
L- Lysine chloride	0.003g
L- Phenyl alanine	0.001g
L- Leucine	0.002g
DL- Tryptophan	0.001g
L- Valine	0.001g
L- Cystein chloride mono hydrate	0.001g

The experimental period was divided into two feeding phases, starter (from 0-3 weeks of age) and grower (from 4-6 weeks of age). The basal experimental diets were described in Table (2). Experimental diets were formulated to meet the nutrients requirements of the broiler chicks according to NRC (1998).

Chicks were full access to feed and water during the experimental period. Artificial light was applied to maintain 23 hrs light per day during the experimental period. The environmental temperature was about 32º C during the first week old and it was gradually reduced by about 2º C weekly until about 24º C at the fourth week up to the end of experiment (at 6 weeks of age). The experiment was included four treatments. Chicks in treatment 1 were not injected with Ca-Aminoplex and served as a control treatment; while chicks in treatment 2 were injected muscularly with 0.3 ml of Ca-Aminoplex at the beginning of the 2nd week, whereas chicks in treatment 3 were injected muscularly with 0.3 and 0.4 ml Ca-Aminoplex at the beginning of 2nd and 4th weeks, respectively. In treatment 4, chicks were injected muscularly with 0.3, 0.4 and 0.5 ml of Ca-Aminoplex at the beginning of 2nd, 4th and 6th weeks of age, respectively. Injection doses were obtained by the calculation involved in the Ca-Aminoplex pamphlet. The calculation is depended on species, age and most correlated with body weight.

Live body weight and feed intake for each replicate were recorded at 0, 1, 2, 3, 4, 5 and 6 weeks of age and average body weight and feed intake was calculated. Feed conversion ratio (gm feed / gm gain) was calculated for each replicate within each period. Dead chicks during the experiment were recorded daily and mortality rate was calculated.

At 6 weeks of age, five chicks from each treatment in the experiment were injected intravenously in the brachial vein with 0.2 ml of 10% suspension of packed sheep red blood cells (SRBC's). Sera were collected on the seventh day post immunization (at 7 weeks of age) and antibody titer against SRBC was determined using the micro titer procedure described by

Van der Zijpp and Leenstra (1980). Titers were expressed as the log2 of the reciprocal of highest dilution giving complete agglutination.

Table 2. Composition of the starter and grower experimental diets.

INGREDIENTS	Starter (from 0-3 wks)	Grower (from 4-6 wks)
Yellow corn	59.00%	65.70%
Soybean meal (44%)	21.00%	17.00%
Corn Gluten (60% CP)	6.66%	4.00%
Broiler concentrate	10.00%	10.00%
Di-Calcium Phosphate	0.50%	0.50%
Lime stone	0.50%	0.50%
Premix	0.25%	0.25%
Lysine	0.18%	0.11%
Methionine	0.41%	0.25%
Vegetable oil	1.50%	1.69%
Total	100.00%	100.00%
Analysis (calculated)		
Crude protein	23.11%	20.33%
ME.	3115%	3162%
Calcium	1.08%	1.07%
Available Phosphorus	0.52%	0.51%
Lysine	1.20%	1.00%
Methionine	0.49%	0.38%
Chemical analysis (determined)		
Crude protein	23.33%	20.42%
Crude fiber	3.51%	3.33%
Crude fat	4.97%	6.68%
Moisture	8.85%	8.89%
Ash	4.98%	4.84%

Cost of one kilogram feed for different diets, cost of feed/kg gain and the cost of feed/bird were calculated on the basis of the price of feed ingredients in the local market. The absolute economic returns were calculated as follow: Selling price/bird and cost of feed/bird. The relative economic returns were calculated in relation to the control treatment.

2- Statistical Analysis:

Data collected were statistically analyzed by the analysis of variance with the General Linear Model (GLM) procedure of the SAS Institute (SAS, 1996). All statements of significance are based on the 0.05 level of probability. Significant differences among treatments were performed using Duncan's multiple range test (Duncan, 1955).

RESULTS AND DISCUSSION

1. Productive performance:

1.1. Body weight:

Data of body weight at 1, 2, 3, 4, 5 and 6 weeks of age are listed in Table (3). Obtained data showed that all treated groups were heavier ($P<0.05$) in body weight at the end of 2nd week than the control group. At 4 and 6 weeks of age, broiler chicks injected two or three times with Ca-Aminoplex, recorded the heaviest ($P<0.05$) body weight than those injected

one time or the control one (not injected). There were insignificant differences in body weight of broiler chicks at 1, 3 and 5 weeks of age. These results partially agreement with Garcia and Mack (2000). Hassan *et al.* (2003) found that, addition of essential amino acids (especially methionine and lysine) improved body weight**.** Amino acids are known to be the building blocks of proteins, so it is expected that the injection mobilize the accretion of amino acids and proteins.

Table 3. Averages ± (SE) of body weight (gm) of broiler chicks as affected by injection with Ca-Aminoplex.

Item / Treatments	Age (week)					
	1	2	3	4	5	6
Control	118.27± 0.64	257.60± 8.59 [b]	525.00± 23.63	861.67± 23.15 [b]	1285.0± 38.84	1470.0± 34.64 [b]
One injection	120.40± 2.75	280.00± 2.89 [a]	530.00± 18.93	843.33± 11.67 [b]	1250.0± 5.77	1431.7± 46.67 [b]
Two injections	126.40± 2.72	282.60± 2.20 [a]	570.00± 7.64	923.33± 10.93 [a]	1315.0± 22.55	1848.3± 26.19 [a]
Three injections	127.13± 2.48	290.40± 3.67 [a]	578.33± 8.82	930.00± 20.82 [a]	1343.3± 26.19	1850.0± 37.53 [a]

Means within each column bearing different letter(s) are significantly (P<0.05)

1.2. Daily gain:

Data of daily gain at 0-3, 4-6 and 0-6 weeks of age are listed in Table (4). Obtained data indicated that the injection with Ca-Aminoplex improved (P<0.01) daily gain during the periods from 4-6 and 0-6 weeks of age. Also, data revealed that, the injected groups with Ca-Aminoplex two or three times recorded higher (P<0.05) daily gain during the periods from 4-6 and 0-6 weeks than the control group and those injected one time. However, the differences were not significant in daily gain between the control group and those injected one time at 4-6 and 0-6 weeks of age. In the starting period, the differences were not significant among different groups. These results are in harmony with Garcia *et al.* (2000) and Abd-Elsamee (2001). Also, El-Husseiny *et al.* (2004) found that, excessive dietary amino acids (methionine and lysine) for the first 7 days of age improved body weight gain. Moreover, excess of vitamins and minerals numerically improved body weight gain.

It is known from Table (1) that Ca-Aminoplex contained some amino acids such as phenyl alanine, lucine, tryptophan and valine. Moreover, it contains vitamins such as vit. B1, vit. B2, vit. B3, vit. B5 and vit. B6. So it is postulated that the Ca-Aminoplex could be improved the daily gain. Thus, it could be recommend injecting broiler chicks with Ca-Aminoplex at least two times to obtain a positive effect on daily gain.

Table 4. Averages ± (SE) of daily gain (gm) of broiler chicks as affected by injection with Ca-Aminoplex.

Item / Treatments	Age (week)		
	0-3	4-6	0-6
Control	22.85± 1.14	45.00± 1.22 [b]	33.92± 0.83 [b]
One injection	23.08± 0.88	42.94± 2.78 [b]	33.01± 1.11 [b]
Two injections	24.98± 0.34	60.87± 0.92 [a]	42.93± 0.62 [a]
Three injections	25.37± 0.43	60.56± 1.55 [a]	42.96± 0.89 [a]

Means within each column bearing different letter(s) are significantly (P<0.05)

1.3. Feed intake:

Data of feed intake at 0-3, 4-6 and 0-6 weeks of age are listed in Table (5). Obtained data illustrated that the injection with Ca-Aminoplex had insignificant effect on feed intake during the starting period. However, during the periods from 4-6 and 0-6 weeks, injection of Ca-Aminoplex increased (P<0.05) feed intake. The results revealed that, the group injected one time ate lower (P<0.05) feed than other groups. Moreover, broiler chicks injected two or three times with Ca-Aminoplex recorded the highest (P<0.05) feed intake during the periods from 4-6 and 0-6 weeks of age. These results are in harmony with Pillai *et al.* (2006) who found that feed intake was maximized (P<0.05) with addition of 0.07% supplemental methionine.

Table 5. Averages ± (SE) of daily feed intake (gm) of broiler chicks as affected by injection with Ca-Aminoplex.

Item / Treatments	Age (week)		
	0-3	4-6	0-6
Control	45.71±0.90	115.08±1.24[b]	80.40±0.88[b]
One injection	47.38±0.60	107.06±0.48[c]	77.22±0.20[c]
Two injections	48.49±0.92	119.60±0.08[a]	84.05±0.48[a]
Three injections	47.78±0.48	118.89±0.69[a]	83.33±0.12[a]

Means within each column bearing different letter(s) are significantly (P<0.05)

1.4. Feed conversion:

Data of feed conversion at 0-3, 4-6 and 0-6 weeks of age are listed in Table (6). Obtained data elucidated that the injection of Ca-Aminoplex had insignificant effect on feed conversion at 0-3 weeks of age, while during the periods from 4-6 and 0-6 weeks of age, injection with Ca-Aminoplex improved (P<0.05) feed conversion. Also, data revealed that, the groups injected two or three times with Ca-Aminoplex recorded the best (P<0.05) feed conversion compared to control group and the group which injected one time during the periods from 4-6 and 0-6 weeks of age. These results are in harmony with Kidd *et al.* (1997). Also, El-Husseiny *et al.* (2004) stated that excessive dietary amino acids (methionine and lysine) for the first 7days of age improved feed conversion and excess of vitamins and minerals numerically improved feed conversion too.

The improvement in feed conversion due to the injection with Ca-Aminoplex can be explained by its composition from several essential amino acids and vitamins which increase the appetite of chicks to raise the feed intake synchronized with rise in metabolism and accretion of amino acids leading to increase the daily gain with optimum utilization of protein. This in good agreement with those obtained by El-Husseiny *et al.* (2004).

Table 6. Averages ± (SE) of feed conversion of broiler chicks as affected by injection with Ca-Aminoplex.

Item / Treatments	Age (week)		
	0-3	4-6	0-6
Control	2.01± 0.11	2.56± 0.08[a]	2.37± 0.06[a]
One injection	2.06± 0.06	2.51± 0.15[a]	2.35± 0.09[a]
Two injections	1.94± 0.04	1.97± 0.03[b]	1.96± 0.02[b]
Three injections	1.88± 0.05	1.97± 0.05[b]	1.94± 0.04[b]

Means within each column bearing different letter(s) are significantly (P<0.05)

1.5. Mortality rate:

There was not any died chick at whole experimental period from any treated group or control group. These mean that viability rates were 100% and the injection with Ca-Aminoplex had no detrimental effect on viability rate.

1.6. Economic revenue:

The economic revenue of broiler chicks injected with Ca-Aminoplex is listed in Table (7). Obtained data illustrated that, the injection with Ca-Aminoplex two or three times decreased ($P<0.05$) the cost of diet/kg gain, but increased the cost of diet/bird at marketing age. On the other hand, broiler chicks injected with Ca-Aminoplex two or three times recorded the highest ($P<0.05$) net revenue and the values ranged from 70.89 and 71.47%, respectively compared to the control group at the end of the experiment (at 6 weeks of age). This could be attributed to the higher body weight gain and the better feed conversion ratio in the injected groups compared to the no injected one. However, broiler chicks, which injected one time and those in the control group, had similar economic value.

2. Immune responses:

Data of serum haemagglutinin antibody (Ab) titers at 7^{th} week of age are listed in Table (8). Obtained data elucidated that injection of Ca-Aminoplex led to significantly ($P<0.05$) effect on Ab. at the 7^{th} week of age. Values of (Ab) titers were higher in the groups injected with Ca-Aminoplex three times followed by those injected two times than the other groups. However, the difference in Ab. between the group injected with Ca-Aminoplex one time and the control group was not significant. These results are in agreement with Edens *et al.* (1999) and Ali (2006). Tryptophan, unlike other single amino acids has been shown to rapidly stimulate long term protein synthesis. The increased anti-SRBC antibody response in the chicks may represent enhanced induction of the immune response via activation of the macrophage system. It is of interest that in the current study, the chicks given a sheep red blood cell antigen challenge and two or three injections of Ca-Aminoplex had higher peak levels of anti-sheep red blood cell antibody than control and one injected groups. This observation suggested that the injection of Ca-Aminoplex in combination with optimum dietary amino acids were sufficient to allow increased protein synthesis without sacrificing growth.

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It is known from Table (1) the composition of Ca-Aminoplex from amino acids such as phenyl alanine, lucine, tryptophan and valine. Moreover, it contains vitamins such as vit. B1, vit. B2, vit. B3, vit. B5 and vit. B6. So it is postulated that the immune response of chicks will be improved as a result of all these important components.

Table 8. Averages ± SE of haemagglutinin antibody (Ab) titers of broiler chicks as affected by injection with Ca-Aminoplex.

Item	Experimental treatments			
	Control	One injection	Two injections	Three injections
Ab. (log_2)	4.28±0.06^{c}	4.25±0.06^{c}	5.20±0.07^{b}	6.23±0.09^{a}

Means within row bearing different letter(s) are significantly (P<0.05)

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تأثيرات الحقن بCa-Aminoplex على بعض الصفات الإنتاجية والاستجابة المناعية لبدارى التسمين

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قسم عدد ١٢٠ كتكوت تسمين غير مجنسين عمر يوم من سلالة الهبرد عشوائيا إلى ٤ مجموعات معاملة. تحتوى كل معاملة على ٣ مكررات بكل مكررة ١٠ كتاكيت (٤ معاملات X ٣ مكررات X ١٠ كتاكيت = ١٢٠ كتكوت).

قسمت مدة التجربة إلي مرحلتين غذائيتين، بادئ (من عمر ٠-٣ أسابيع) ونامى (من عمر ٤-٦ أسابيع). وكانت علائق التجربة الأساسية تحتوى على ٢٣,٣٣ و ٢٠,٤٢% بروتين خام و ٣١١٥ و ٣١٦٢ كيلو كالورى طاقة ممثلة/كجم عليقة لكل من عليقتى البادئ والنامى على الترتيب. تضمنت التجربة ٤ معاملات كالتالى: فى المعاملة الأولى لم يتم حقن الكتاكيت بمركب Ca-Aminoplex واستخدمت كمجموعة مقارنة، بينما تم حقن كتاكيت المعاملة الثانية عضليا ب٠,٣ مل من ال Ca-Aminoplex فى بداية الأسبوع الثانى، بينما تم حقن كتاكيت المعاملة الثالثة عضليا بنفس المحلول بمستوى ٠,٣ و٠,٤ مل فى بداية الأسبوع الثانى والرابع على الترتيب. أما فى المعاملة الرابعة فقد تم حقن الكتاكيت عضليا من نفس المحلول ب ٠,٣ و ٠,٤ و ٠,٥ مل فى بداية الأسبوع الثانى و الرابع و السادس على الترتيب. تم الحقن الوريدى لـ٥ كتاكيت من كل معاملة بمحلول كرات الدم الحمراء للغنم (٠,٢ مل) وتم جمع السيرم فى اليوم السابع من الحقن لتقدير مستوى إنتاج الأجسام المناعية ضد كرات الدم الحمراء للغنم.

ويمكن تلخيص النتائج فى الاتى:

أظهرت البيانات المتحصل عليها أن كل المجموعات المحقونة بالCa-Aminoplex كانت أعلى (P<0.05) معنويا فى الوزن فى نهاية الأسبوع الثانى عن مجموعة المقارنة، بينما كانت المجموعتين المحقونتين مرتين أو ثلاث مرات بCa-Aminoplex أعلى معنويا (P<0.05) فى الوزن فى نهاية الأسبوعين الرابع والسادس عن مجموعة المقارنة والمجموعة المحقونة مرة واحدة.

أدى حقن الكتاكيت مرتين أو ثلاث مرات ب Ca-Aminoplex إلى زيادة معدل الزيادة اليومية فى وزن الجسم واستهلاك الغذاء اليومى (P<0.05) وتحسين معدل التحويل الغذائى (P<0.01) عند ٤-٦ و٠-٦ أسبوع من العمر بالمقارنة بمجموعة المقارنة والمجموعة المحقونة مرة واحدة بالCa-Aminoplex.

أدى حقن الكتاكيت مرتين أو ثلاث مرات بالCa-Aminoplex إلى انخفاض تكلفة الغذاء لكل كجم زيادة فى الوزن وزيادة تكلفة الغذاء/ طائر وزيادة العائد الصافى بالمقارنة بالكنترول أو المجموعة المحقونة مرة واحدة بالCa-Aminoplex.

كان هناك تأثير معنوى (P<0.05) للحقن بCa-Aminoplex على الأجسام المضادة لكرات الدم الحمراء للغنم بسيرم الدم فى الأسبوع السابع حيث كانت أعلى فى المجموعة المحقونة ثلاث مرات عن باقى المجموعات، علاوة على ذلك فان المجموعة المحقونة مرتين كانت أعلى فى كمية الأجسام المضادة لكرات الدم الحمراء للغنم عن مجموعة المقارنة والمجموعة المحقونة مرة واحدة.

من ذلك يمكن القول بان حقن الكتاكيت بCa-Aminoplex مرتين أو ثلاث مرات يؤدى إلى تحسين أداء الكتاكيت الانتاجى والاستجابة المناعية لها.

Table 7. Economic revenue of broiler chicks as affected by injection with Ca-Aminoplex.

Items / Treatments	Price of diet/kg		Cost of diet/kg gain			Cost of diet/bird			Cost of aminoplex /bird	Bird price	Net revenue	% From control
	Starter	Grower	Starter	Grower	*Total*	Starter	Grower	Total				
Control	1.96	1.81	3.94	4.64^{a}	4.29 ^{a}	1.88	4.37^{b}	6.26^{b}	0	9.04^{b}	2.78^{b}	100.00
One injection	1.96	1.81	4.03	4.55^{a}	4.29 ^{a}	1.95	4.07^{c}	6.02^{c}	0.03	8.80^{b}	2.75^{b}	98.78
Two injections	1.96	1.81	3.81	3.56^{b}	3.68 ^{b}	2.00	4.55^{a}	6.54^{a}	0.07	11.37^{a}	4.76^{a}	170.89
Three injections	1.96	1.81	3.69	3.56^{b}	3.63 ^{b}	1.97	4.52^{a}	6.49^{a}	0.12	11.38^{a}	4.77^{a}	171.47

Means within each column bearing different letter(s) are significantly (P<0.05)

-Selling price was 6.15 LE/kg.

-Cost of diet/kg gain = feed conversion x cost of kg feed.

-Total cost of diet/kg gain= (feed conversion during starting period x cost of kg starter diet + feed conversion during growing period x cost of kg grower diet)/2.