CONTENT OF GLYCOGEN IN LIVER AND KETOBODIES IN BLOOD OF JAPANESE QUAILS (COTURNIX COTURNIX JAPONICA) DURING STARVATION ENVISAGED IN THE METHODS OF BALANCE EXPERIMENTS

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ABSTRACT

The biochemical indexes “glycogen in liver” and “ketones in blood” of 0-72 hours feed deprived (according to methods for balanced experiments) Japanese quails with and without energy additives were determined.

There were 2 groups of birds- 1-st without energy supplement, 2-nd- fed with 1 g. glucose per os (as 25% solution) – twice in 24 hours.

The levels of liver glycogen in all the food-deprived quails were significantly lower from -6910 (12-th hour of starving)-to 4960mg/kg (72 hour of starving) compared to the levels of the same index in fed birds (11990 mg/kg tissue). In the birds receiving energy additive they were higher compared to those deprived of the additive throughout the experimental period.

The content of ketones in blood of the control birds was 0.015 mmol/l. The same index increased to 0.027 mmol/l in the feed and energy additive deprived group after the 36 hour of starving, but in the group became energy support, the contents of ketones were lower for the whole period of starving.

The energy additive (1g glucose/24 hours) helped the maintenance of the energy metabolism during continuous food depriving of the experimental quails.

KEYWORDS: Glycogen, Liver, keto- bodies, Blood, Japanese quails

РЕЗЮМЕ

Установени са биохимичните показатели “гликоген в черния дроб “ и “кетони в кръвта” при 0-72 часа лишени от храна Японски пъдпъдъци с и без енергийна добавка (съгласно методика за балансови опити).

Формирани бяха 2 групи от птици – първата без енергийна добавка, а втората получаваше по 1g глюкоза per os (като 25% р-р) – двукратно в денонощието.

Нивото на чернодробния гликоген в черния дроб на всички лишени от храна пъдпъдъци беше значително по-ниско -от 6910 (на 12 час от гладуването) – до 5690mg/kg (на 72 час от гладуването) в сравнение със същия показател при хранените птици (11 990mg/kg). При птиците получавали енергийна добавка, то бе по-високо в сравнение с неполучавалите такава за целия опитен период.

Съдържанието на кетотела в кръвта на контролните птици бе 0,015 mmol/l. Същия индекс нараства до 0,027mmol/l при гладуване и лишаване от енергийна добавка след 36 час, но при групата с енергийна добавка съдържанието на кетони бе по-ниско за целия опитен период.

Енергийната добавка (1g глюкоза / 24 часа) подпомага поддържането на енергийния метаболизъм при продължително лишаване от храна на опитните пъдпъдъци.

КЛЮЧОВИ ДУМИ: Гликоген, черен дроб, кетони, кръв, Японски пъдпъдъци

ПОДРОБНО РЕЗЮМЕ

Правилното провеждане на балансови опити с птици изисква възможно най-пълно очистване на храносмилателния тракт от фуражни остатъци. Това се постига чрез лишаване от храна за определено време. При птиците този период е с продължителност от 48-108 часа. Енергийния метаболизъм се повлиява при гладуването и телесните резерви се мобилизират. Това трябва да се установи много прецизно за максимална компенсация на отрицателния енергиен баланс на птичия организъм.

Не се намериха данни за съдържанието на гликогена в черния дроб и кетотела в кръвта на пъдпъдъци в достъпната ни литература.

Целта на настоящето проучване е да се установи динамиката на количествените промени на гликогена в черния дроб и ацетонови тела в кръвта на пъдпъдъци, лишени от храна според методиката за балансови опити с птици.

През 2004 г. проведохме опит с 78 Японски пъдпъдъци от породата Фараон на 4-5 седмична възраст от едно и също люпило/ по 36 във всяка опитна група и 6 птици в контролната група. Опитните групи бяха сформирана по седния начин: Първата опитна група беше лишена от енергийна добавка за 72 часа. Втората опитна група получаваше по 1g чиста глюкоза като 20% разтвор на всеки 12 часа за същия период на лишаване от храна.

Съдържанието на гликоген беше определено чрез модифицирана методика комбинирана с тест Glucose GOD FS* - Diagnostic Systems – Germany.

Кетотелата (в mg ацетон/1000 ml проба) бяха определяни фотометрично по модифицирана методика на Natelsson.

Енергийната добавка (1g чиста глюкоза на 24 часа бе извършена според данните за необходимата нето енергия за поддържане при птици.

Еднофакторния дисперсионен анализ бе използван за сравняване на резултатите, като достоверността на разликите определихме по Стюдънт – Фишер.

Съдържанието на гликоген в черния дроб на птиците от контролната група бе 11990 ± 96 mg/kg. При гладуване молекулата на гликогена е подложена на бързо разграждане, при което се освобождава глюкоза, осигуряваща енергия. При птиците, лишени от енергийна добавка стойностите на този индекс намалят от 12 час от гладуването (6910 ± 160 mg/kg), като тенденцията се запазва до 24 час (2740 ± 230 mg/kg), ($p \leq 0.01$). Различията с контролната група, макар и недостоверни, остават същите докрая на опитния период.

Средното съдържание на кетотела в кръвта на негладували пъдпъдъци бе 0.014764±0.005 mmol/l. При гладувалите пъдпъдъци, но нетретирани с глюкоза стойностите на индекса варираха без ограничение, 0.001188±0.00005 до 0.026618±0.001 mmol/l. По-ниско ниво на кетотелата в сравнение с контролните бяха на 12 час (0.001188±0.00005), а с най-висока степен на достоверност на 36 и 48 час от началото на опитния период.

Енергийната добавка от 1g чиста глюкоза подпомага поддържането на енергиййния метаболизъм при продължително лишаване от храна на опитните Японски пъдпъдъци.

INTRODUCTION

The proper carrying out of balance experiments with birds requires the best possible clearing of the digestive tract from forage residues. That is achieved by food depriving for a certain time. For fowls those periods were in the frames of 48 – 108 hours (16; 1; 13; etc.).

The abrupt change of the nutrition regime caused stress in the organism, especially in the high productive bird races (3). During food depriving the content of liver glycogen can be totally exhausted. The glycogen in muscles under the same conditions is usually not exhausted to drastically low levels. The energy metabolism is affected during starvation and body reserves are being mobilized. That should be established very precisely in order to compensate the negative energetic balance in the bird organism to the maximum. Besides, the results of the experiment should be comparable with those in practice, i.e. they should serve the aims of nutrition in practice.

The unfavourable effect of the negative energetic balance on the growth and development of the young organisms is well known. For that reason (9) suggested that the food-deprived, 22-24-week old Peking ducks submitted to balance experiments should be additionally given 30 % solution of glucose from 8th until 60th hour of starvation. It is practically 100 % absorbable and does not affect the results of digestibility.

During total or partial food depriving the amount of glycogen in liver and muscles decreased significantly due to its intensive disintegration (15).

Ketobodies are normal products of the metabolism of fats and carbohydrates. They were found in minimal amounts in the blood of domestic animals (7).

We did not find data about their amounts in the blood of quails in the available literature.

The aim of the present study was to establish the dynamics of the quantitative changes of the glycogen in liver and of ketobodies in the blood of quails food-deprived in accordance with the methods of balance experiments with fowls (13).

MATERIAL AND METHODS

In 2004 experiments were carried out with 78 Japanese quails of Pharaon breed at the age of 4-5 weeks of the same hatching /by 36 in each experimental group/ and 6 birds in the control group. The experimental groups were formed in the following way: The first experimental group was deprived from the energy additive for the whole period of food depriving lasting for 72 hours. The second experimental group received by 1 g of pure glucose as 20 % water solution twice every 12 hour for the same period of starvation. The following indices were reported for the control group fed on standard forage mixture, and, for the experimental groups: liver glycogen and ketobodies in the peripheral blood. In both experimental groups those two indices were tested by equal 12-hour intervals until the end of the investigation – the 72nd hour. Six quails were used for each test. Glucose was inserted twice by 12-hour interval per os as 20 % solution, the total dose being 1 g of pure glucose for 12 hours. The dynamics of the ketobodies was determined by puncture of v. jugularis and the content of glycogen in liver – from lobus caudatus – detected immediately post mortem, strictly following the internationally adopted standards in that sphere.

The glycogen amount was determined by the modified methods of (8) combined with the test Glucose GOD FS* - Diagnostic Systems – Germany.

Ketobodies (in mg of acetone/1000 ml sample) were determined photometrically by modified methods of Natelsson (2).

The energy additive (1 g of pure glucose per 24 hours) was determined by the data about the necessary net energy for breeding birds (12).

Monofactor dispersion analysis was used for comparing the obtained results, the significance of the differences being calculated by Student – Fischer (18).

RESULTS AND DISCUSSION

Table 1 presents the dynamics of glycogen in quail liver.

Glycogen content in the birds of the control group was 11990 ± 96 mg/kg. Our results were compatible with those of (17) and (4). During starvation the glycogen molecule was submitted to rapid disintegration, releasing glucose that supplied energy. Due to its intensive disintegration the amount of glycogen in liver and muscles decreased significantly (15). Liver glycogen was easily mobilized when the energy requirements of the organism were high (19, 5, 11, 10, 4).

Table 1. Content of glycogen in Japanese quail's liver (mg/kg).

Feed depriving – hours	12			24			36			48			60			72		
Control group	11990± 96 / 21.28 a, a1, a2, nq																	
	x	Sx	S%	x	Sx	S%	X	Sx	S%	X	Sx	S%	X	Sx	S %	X	Sx	S%
Feed deprived quails without energy supply	6910 a, nq1	160	3.99	2740 a,b	230	14.53	3770 b, nq	180	8.18	6340 b, nq	750	20.58	6600 nq, nq1	160	4,96	5690 nq, nq1	1350	47,59
Feed deprived quails with supply 1 g glucose twice in 24 hours	17440 a1, nq, nq1	1370	15.77	16920 a2, b	830	9.80	13880 b, nq	500	6,26	15620 a2, b	1100	1,18	15110 nq, nq1	490	6,43	9640 nq, nq1	620	12.78

Legend: Statistical significance: nq-nq- no, between control and the groups, nq1-nq1- no between 1-st and 2-nd group, a-a – significant by p< 0.01 between control and 1-st group; a1-a1 - significant by p< 0.1 between control and 2-nd group; a2-a2 - significant by p< 0.05 between control and 2-st group; b-b - significant by p< 0.01 between 1-st and 2-nd group

Fig. 1. Content of glycogen in Japanese quail's liver after starving (mg/kg).

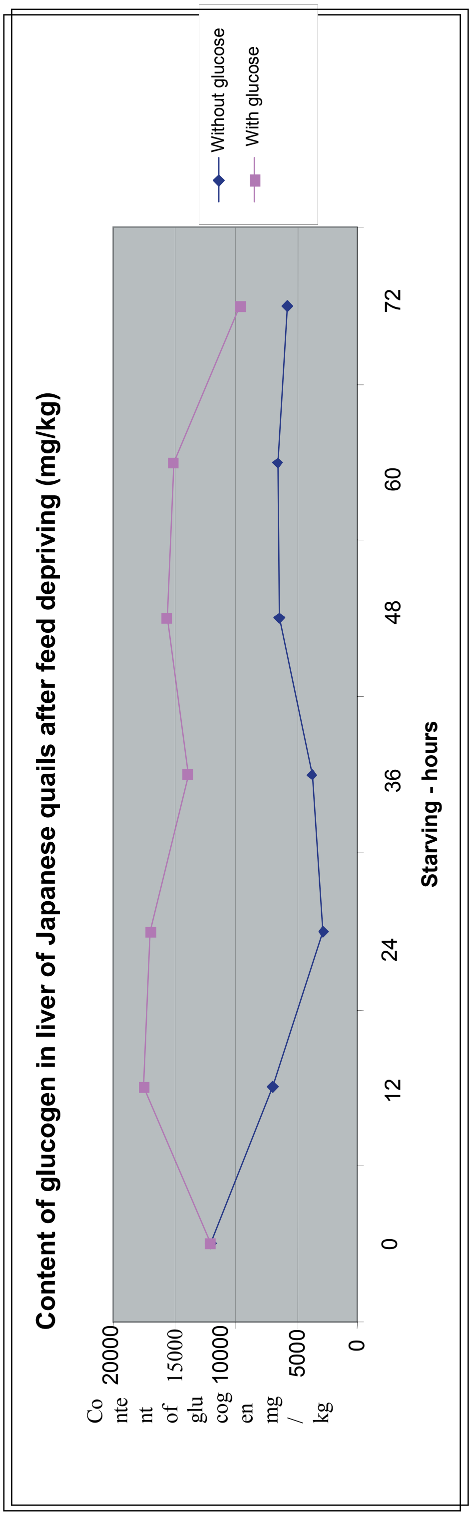

Fig. 2. Content of ketones in Japanese quail's blood after starving (mmol/l).

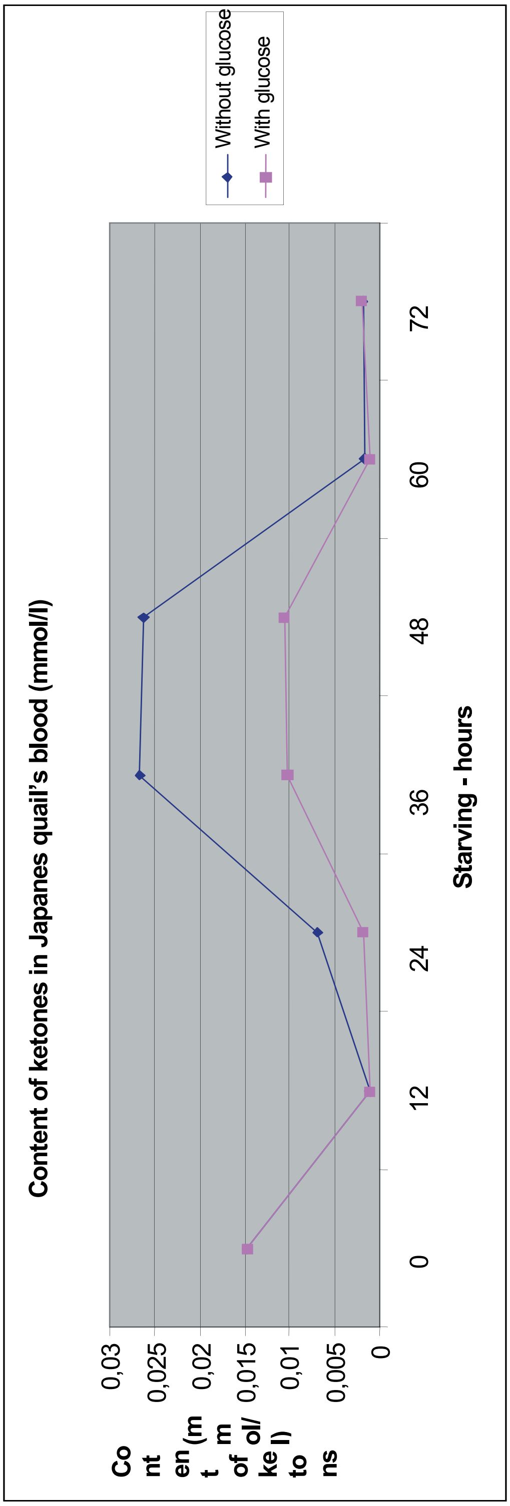

Table 2. Content of ketones in Japanese quail's blood (mmol/l).

Feed depriving – hours	12			24			36			48			60			72		
	x	Sx	S%	X	Sx	S%	x	Sx	S%	x	Sx	S%	X	Sx	S%	X	Sx	S%
Control group	0.014764±0.005/119.02 a, a1, nq																	
Feed deprived quails without energy supply	0.001188 b, nq	0.00005 a,b	9.20	0.006832 a, b	0.002	87.14	0.026618 a,b	0.001	103.27	0.026268 a,b	0.002	12.96	0.001592 a,b	0.001	81.93	0.001871 a,b	0.001	66.77
Feed deprived quails with supply 1 g glucose twice in 24 hours	0.001162 a1, b, nq	0.001	41.55	0.001837 a1, b	0.001	70.50	0.010244 a1, b	0.005	127.14	0.010670 a1, b	0.002	43.66	0.001080 a1, b	0.000	75.13	0.002135 a1, b	0.000	42.36

Legend: Significance: nq-nq- no; a-a - significant by p< 0.01 between control and 1-st group;
a1-a1 – significant by p< 0.01 between control and 2-d group; b-b - significant by p< 0.01 between 1-st and 2-nd group

In the birds deprived of energy additives the values of that index decreased significantly as early as the 12th hour of starvation (6910 ± 160 mg/kg), the tendency being maintained until the 24th hour (2740 ± 230 mg/kg), (p≤0.01). The differences to the control group, although insignificant, remained the same until the end of the experimental period (Table 1). Figure 1 presents graphically the glycogen levels in the food deprived birds with and without an energy supply with glucose solution. It should be mentioned that the levels of liver glycogen in all the quails were lower than the levels of the same index in the non-starving birds of the control group.

The levels of liver glycogen in the birds that received glucose solution were significantly higher at the 24th, 36th and 48nd hour (p≤0.01) in comparison with the quails without the energy additive (Table 1, Figure 1).

Table 2 presents the dynamics of ketobodies in the blood of the studied quails.

The mean values of ketonic bodies in the blood of the non-starving quails were 0.014764±0.005 mmol/l. Our results were the closest to those cited about pigs (7, 6). In the available literature we did not find data about the levels of that index for quails. In our previous studies with young Muscovy ducks (14) the values of the ketonic bodies in the peripheral blood of non-starving birds were 0.0095 mmol/l after which they increased significantly until the 72nd hour, decreasing afterwards.

In the untreated but food-deprived quails the values of the index varied within the limits of 0.001188±0.00005 to 0.026618±0.001 mmol/l. The lowest levels of ketobodies compared to the control were at the 12th hour (0.001188±0.00005), and the highest statistically significant levels were at the 36th and 48th h after the beginning of the experiment (Fig. 2).

The acetone bodies in the second experimental group were lower compared to the first one and they varied within narrower limits during the whole period of the investigation.

In the first experimental group (starving quails without glucose additive) a high degree of significance was detected in the dynamics of content of ketobodies, determined by the duration of starving (Table 2). The content of ketonic bodies in the second experimental group (starving quails with glucose additive) followed the dynamics of the index in the first experimental group but the values were significantly lower (with an exception of the period 60th – 72th hour (Table 2, Fig. 2).

Total acetone bodies are organic compounds containing: acetone, acetoacetic acid and β – oxyfat acid. They are normal metabolites and they are to be found in minimal quantities in the blood of pigs and ruminants – 0.5 - 8.0

mg %, (6, 7, etc.). In human peripheral blood they are below 30 mg/l (15). Data of the present experiments were close to those cited about pigs.

CONCLUSIONS

The content of glycogen in quail liver varied from 16920 mg/kg (for quails treated with glucose at the 24th hour) to 2740 mg/kg for quails untreated with the energy additive at the 24th hour of the period of starvation. The values of liver glycogen were significantly lower in all the starving quails not receiving glucose compared to the same index for the non-starving birds. As a whole the levels of liver glycogen were significantly higher in the birds receiving glucose in comparison with those deprived of the energy additive.

The content of ketones in the bird blood was the lowest for both experimental groups at the 12th h of the period of starvation (0.00117mmol/l) and the highest – at the 36th h of the period of starvation - 0.010 mmol/l in the quails receiving glucose and 0.027 mmol/l in those deprived of the glucose, respectively. The energy additive given to the starving quails maintained relatively stable levels of acetone bodies in blood. The ketonic bodies in the blood of the starving quails that were given the energy additive were comparatively lower than the level in the starving analogues untreated with glucose.

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