The Research of Medical Science Review
ISSN: 3007-1208 & 3007-1216 Volume 4, Issue 4, 2026

EFFECTS OF HIGH-PROTEIN AND HIGH-CARBOHYDRATE DIETS ON HEMATOLOGICAL, BIOCHEMICAL, AND GLUCOSE PROFILES IN RABBITS

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DOI: https://doi.org/10.5281/zenodo.19704690

Keywords
Rabbits, High-protein diet, High-carbohydrate diet, Hematology, Biochemistry, Glucose

Article History
Received: 28 February 2026
Accepted: 07 April 2026
Published: 23 April 2026

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Abstract
An experiment was conducted to evaluate the effect of high protein and high carbohydrate diets on blood clinical and glucose values in rabbits. Male domestic rabbits (n=18) were purchased from the local market of Hyderabad and brought to the animal house at FAHVS, SAU Tandojam. Rabbits were assigned to three groups: Group A (control), Group B (85% carbohydrate) and Group C (65% protein). They were placed in a controlled, age-appropriate environment with unrestricted access to feed and water. The experimental trial was held for 6 weeks. The results revealed that Group C (10.98±0.36) had consistently higher numbers of RBCs than Group A (7.03±0.46) and Group B (5.037±0.33). WBC count was significantly elevated in Group B (23.060±1.0126) than in Group A (9.742±0.29) and Group C (8.60±0.45). Hb count was significantly elevated in Group C (9.87 3± 0.37) than in Group A (8.298± 0.943) and Group B (5.825± 0.081). ALT count was significantly elevated in Group C (118.00±4.27) than in Group A (39.33±3.32) and Group B (73.00±4.48). AST count was significantly elevated in Group B (121.17±5.49) than in Group A (47.00±2.21) and Group C (67.18±3.52). GGT count was significantly elevated in Group C (14.00±1.46) than in Group A (4.10±0.27) and Group B (11.00±0.97). Total protein (TP) count was significantly elevated in Group C (10.747 ±0.20) than in Group A (4.5333±0.42) and Group B (2.2583±0.863). Albumin count was significantly elevated in Group C (7.2067±0.2557) than in Group A (3.4333±0.2578) and Group B (0.2900±0.0347). Globulin count was significantly increased in Group C (7.2161±0.2659) compared to Group A (3.4423±0.2776) and Group B (0.2800±0.0345). Glucose count was significantly elevated in Group B (277.17 ± 20.51) than in Group A (92.33 ± 3.48) and Group C (10.33 ± 8.59). Total cholesterol count showed no significant differences between the groups, with Group A (74.167±2.77) being slightly higher than Group B (12.00±1.18) and Group C (19.00±1.59). HDL-Cholesterol count was significantly elevated in Group C (29.00±1.32) than in Group A (4.80±0.14) and Group B (16.00±1.65). LDL-Cholesterol count was significantly elevated in Group C (34.00±3.12)

than in Group A (32.00±0.97) and Group B (4.50±0.84). Triacylglycerol count was significantly elevated in Group B (27.83±3.10) than in Group A (10.28±0.66) and Group C (10.28±0.66). Bilirubin count was significantly elevated in Group B (0.8400±0.0603) than in Group A (0.8400±0.0603) and Group C (0.2800±0.0493). The results suggest that elevated levels of carbohydrates and protein have a positive impact on the general well-being of rabbits. This was evidenced by alterations in the CBC count, elevation of LFTs due to high protein intake, modification of the lipid profile, and an increase in blood glucose values.

INTRODUCTION

Rabbits are tiny animals that belong to the family Leporidae and the phylum Lagomorpha. several locations around the world. Meadows, woods, forests, and grasslands all serve as their habitats. The rabbit is widely bred, extremely docile, non-aggressive, and simple to handle and observe. incredibly affordable when compared to the cost of larger animals. Rabbits' life cycles are brief. (Elsheikh et al., 2023). (Gestation, breastfeeding, and puberty). Rabbits are thought to be better because of their great proficiency and rapid growth; it is more efficient than other livestock (Sharma & Choudhary, 2017).

Both their production capacity and feed conversion efficiency are good in contrast to other meat-producing animals. Rabbits are slaughtered between 18 and 20 weeks of age, when they weigh 3 kg under typical circumstances. Rabbit meat is exceptionally flavorful, low in fat and cholesterol (4%), easy to digest, rich in protein content (25%) and low in calories (160 Kcal/100 g of meat). As a result, both children and those with heart conditions can ingest it. Humans have used rabbits for various purposes throughout history because they are intriguing and versatile animals. Here are a few of rabbits' typical uses and functions, including food, fur, study, and pets. They possess the exceptional capacity to procreate and expand quickly while consuming diets higher in fibre and lower in grains year-round (Para et al., 2015). Green grasses, which are distinctive to their diet and easy to digest, are the food they eat. Instead of a more stagnant and contaminated diet, they choose to consume fresh plant components that are nutrient-dense, like leaves and shoots (Queensberry & Orcutt, 2012). Therefore, because they naturally prefer a diet with adequate energy density, rabbits are known as concentrates favoured because the concentrated feed predisposes them to fatness in confinement (Meredith, 2015). According to their anatomical structure, rabbits are simple stomach herbivores, and fermentation occurs in their intestines. The cecum, a region of the hindgut, contains colonies of microbes that aid in digesting nutrients absorbed in the small intestine (Mayer et al., 2017). The relationship between food and cecocolic motility, and the health of cecal bacteria, is essential. A complex milieu of microorganisms, including huge anaerobic metachromatic-staining bacteria, Bacteroides species, and several as-yet-unidentified species of bacteria, may be identified in the cecum, which acts as a fermentation chamber. The bacterial colonies present in the cecum are mostly gram-positive Bacteroides spp. Therefore, due to this the rabbit becomes very sensitive to oral antibiotics, a lot of administration of oral antibiotics can disturb the Bacteroides population and may lead to fatal GI upsets (Heczko Proença et al., 2000; Proença et al., 2014). In the hindgut, digests are divided according to particle size. Large (>0.5 mm) particles, mostly lignocellulose, are swiftly transported down the colon by peristaltic activity, where they are excreted as hard faecal pellets, referred to as "indigestible fibre" in the diet (Gidenne et al., 2020). The therapeutic value of a diet rich in long particle length is to preserve the colon and cecum. This is why these fibres are frequently referred to as the "scratch factor," since they manually enhance GI motility (Oglesbee & Jenkins, 2012; Kubkomawa, 2019). The antimotility function propels tiny particles (< 0.3 mm) and dissolved substances The rabbit may occasionally expel its cecal contents as "soft faeces" or Cecotroph, which it then consumes directly from its anus. Rabbits consume the most cecotrophs when given a diet with a high amount of fibre that cannot be easily digested (Gidenne et al., 2020; McNitt et al., 2013) into the cecum, where they are subjected to fermentation. This component of the diet is known as "digestible" or "fermentable" fibre (Hulls, 2015).

The re-consumed substance provides crucial microbial protein, as well as all the necessary B vitamins and trace amounts of volatile fatty acids that are vital for the well-being of rabbits. The amino acids obtained in this way do not contribute much to the protein needs of rabbits, especially young and growing ones. Therefore, the diet should provide additional amino acids, but rabbits' essential amino acid requirements have not yet been determined (Mayer et al., 2017).

The protein and carbohydrate needs of rabbits for their diet are influenced by several factors, including age, physiological state, environmental conditions, and health. Growth, reproduction, lactation, and body tissue preservation require protein. Rabbits rely on carbohydrates as their primary energy source, which can be divided into two categories: structural (fibre) and non-structural (starch and sugars). (Belenguer et al., 2012). Maintaining normal gut function and preventing digestive disorders such as enteritis, ileus, and cecal dysbiosis greatly depends on the role of fibre (Ehmke et al., 2016). Though rabbits can obtain readily available energy from starch and sugars, excessive intake can result in obesity, dental problems, and metabolic disorders (Ehmke et al., 2016). The appropriate amounts of protein and carbohydrates for rabbits' diets have not been definitively determined.

There are conflicting reports on the effects that high protein and high carbohydrate diets have on rabbits' clinical blood values and glucose levels. There have been several studies conducted on the effects of high-protein diets on blood parameters. While some studies suggest that such diets could have a positive impact on parameters like haemoglobin, haematocrit, total protein, albumin, globulin, urea nitrogen, creatinine, and glucose, others have found that they could have negative effects on parameters like cholesterol, triglycerides, alkaline phosphatase, aspartate aminotransferase, alanine aminotransferase, lactate dehydrogenase, and insulin. On the other hand, studies have shown that high-carbohydrate diets could improve blood parameters such as glucose, insulin, glucagon-like peptide-1, and peptide YY. (Harcourt-Brown et al., 2020; MSD Veterinary Manual, 2021). Some research has suggested that diets high in carbohydrates may hurt various blood parameters, including cholesterol, triglycerides, free fatty acids, beta-hydroxybutyrate, leptin, adiponectin, and resistin (Varga, 2014; Veterinary Practice, 2020).

Further research is necessary to investigate the effects of high protein and high carbohydrate diets on clinical blood and glucose values in rabbits. The purpose of this study is to compare three different diets, including a control diet with normal levels of protein and carbohydrate, a high protein diet with increased protein levels (65%), and a high carbohydrate diet with increased carbohydrate levels (85%), to determine their impact on these parameters. The hypotheses are that the high-protein diet will enhance clinical blood and glucose values, while the high-carbohydrate diet will hurt clinical blood clinical and glucose values. The results of this study will aid in the comprehension of rabbit nutritional physiology and the creation of optimal diets for pet rabbits.

MATERIALS AND METHODS

3.1 Study Area

The study was carried out at the Animal House, Faculty of Animal Husbandry and Veterinary Sciences, Sindh Agriculture University Tandojam. The animals were assigned to different experimental groups randomly, based on the various treatments.

3.2 Experimental design

During the study, 45 rabbits were randomly assigned to three groups, with each group consisting of 15 rabbits (n=15). The first group, which was the control group, did not receive any protein or carbohydrate supplementation, while the second group was given protein supplementation along with their regular diet.

The third group was given carbohydrate supplementation. The study lasted for 42 days.

Table 3.1 Experimental Design

Group A control	Group B high carbohydrate	Group C high protein
15 rabbits	15 rabbits	15rabbits

Animal Care: The rabbits were kept in cages on an individual basis and given free access to water, along with a diet consisting of pellets.

3.3 Dietary Formulation and Feeding Protocol

The study involved formulating three different diets: the Control Diet (CD), the High Protein Diet (HPD) with 65% protein, and the High Carbohydrate Diet (HCD) with 85% carbohydrate, to meet the nutritional requirements of rabbits. All the diets had equivalent energy content, and rabbits in each group were fed their respective diets for six weeks. The feeding process was carefully monitored to ensure that each rabbit consumed the appropriate amount of food.

Table 3.2 Feed Formulation

Dietary component	Group A (control) Fed with Basal diet %	Group B (treated with high carbohydrate diet) 85%	Group C (treated with a high-protein diet) 65%
Maize	32	65	20
Wheat bran	28	15	22
Fish meal	18	10	45
Soybean meal	15	5	8
Mineral mixture	2	2	2
Amino acids	1	2	2
Sunflower oil	1	1	1
Total	100	100	100

3.4 Data Collection

At the beginning of the study, the clinical and glucose values of the blood were measured and recorded as baseline measurements. Throughout the study, the relevant parameters were periodically measured by collecting blood samples.

3.5 Blood Parameters Measurement

The standard laboratory techniques were employed to measure the levels of red blood cell count, white blood cell count, and haemoglobin. Additionally, liver function parameters such as ALT, AST, GGT, TP, albumin, and bilirubin were examined to evaluate liver health. Furthermore, blood glucose levels were assessed in a fasting state.

3.6 Statistical analysis

The "Student Edition of Statistics" computer program was utilized to perform a statistical analysis of the data and determine the coefficient of correlation and regression between the parameters. Results were deemed significant at a P-value < 0.05.

RESULTS

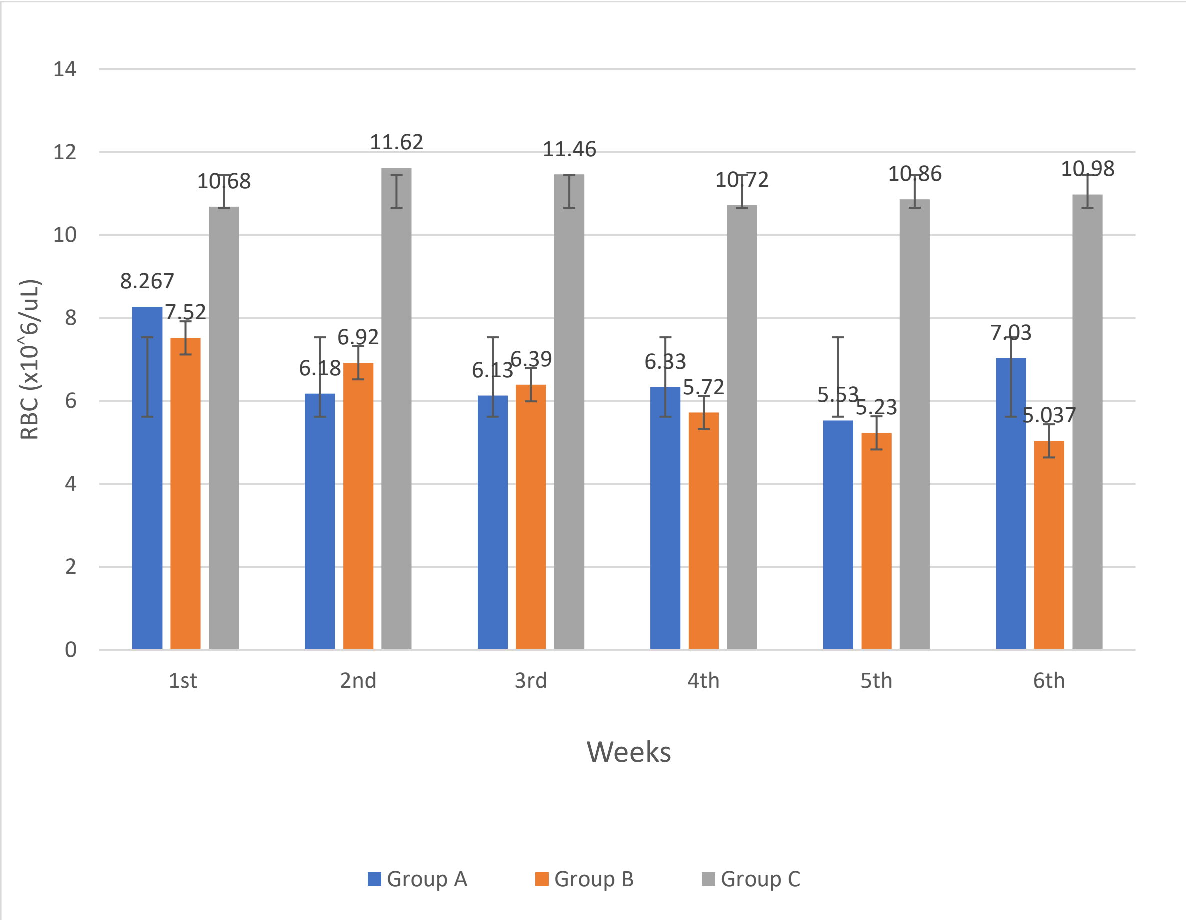
1 Effect of different diets on the red blood cell (RBC) count in rabbits

The effect of different diets on the number of red blood cells in rabbits over six weeks is shown in Table 1&Fig. 1. The differences between the three groups were not statistically significant in any week ($P>0.05$). The initial number of red blood cells for all groups was similar. However, over the six weeks, Group C (fed with a diet containing 65% protein enriched with vitamins and minerals) had consistently higher numbers of red blood cells than Group A (fed with a normal diet) and Group B (fed with an 85% Carbohydrate). At the end of the sixth week, the number of red blood cells in rabbits was slightly higher (10.98±0.36 million per microliter) in Group C than in Group A (7.03±0.46 million per microliter) and Group B (5.037±0.33 million per microliter), but the difference was not significant.

Table-1 Effect of different diets on the red blood cell (RBC) count in rabbits

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
RBC	1st	8.267±0.39	7.52±0.45	10.68±0.48	0.908
	2nd	6.18±0.38	6.92±0.46	11.62±0.49	0.870
	3rd	4.13±0.30	6.39±0.42	11.46±0.33	0.775
	4th	6.33±0.32	5.72±0.41	10.72±0.26	0.635
	5th	5.53±0.38	5.23±0.35	10.86±0.49	0.728
	6th	7.03±0.46	5.037±0.33	10.98±0.36	0.731

Fig- 1 Effect of different diets on the red blood cells (RBC) count in rabbits



2 Effect of different diets on the white blood cell (WBC) count

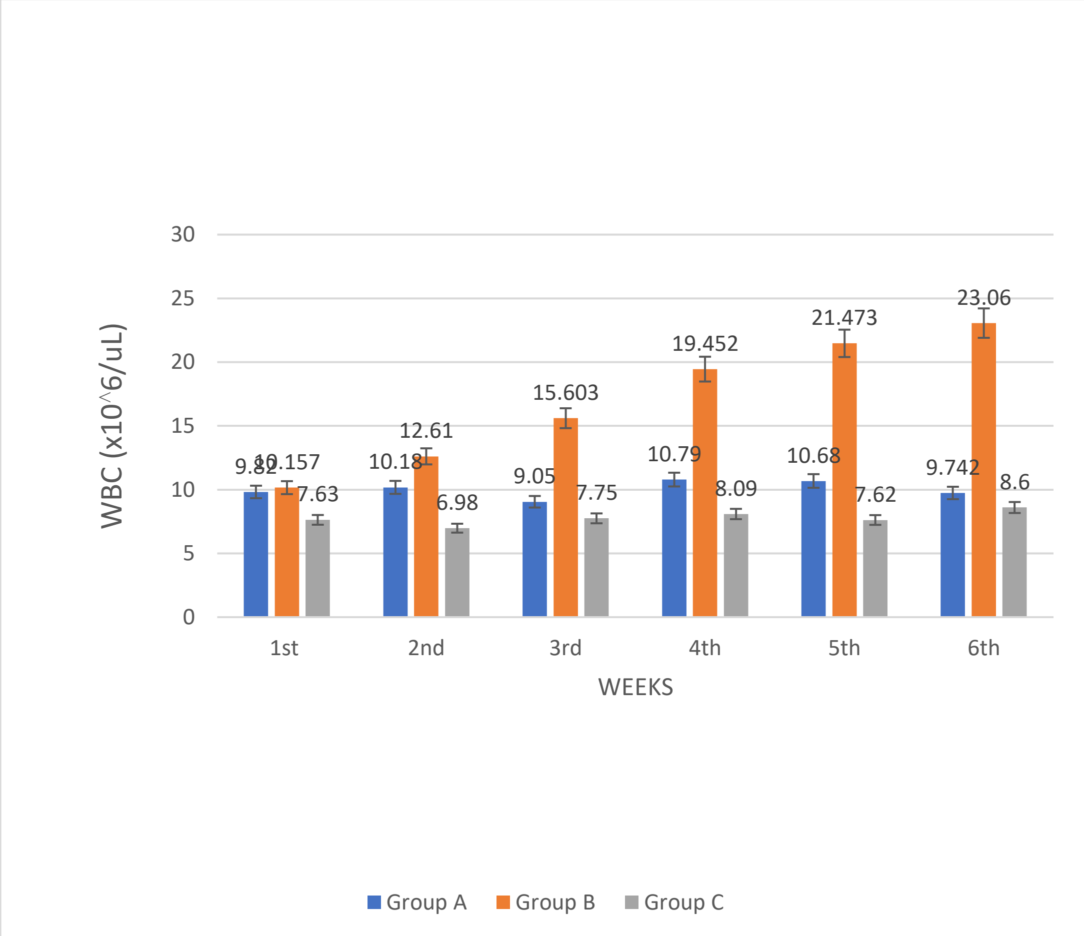
The effect of different diets on the number of white blood cells (WBC) in rabbits over six weeks is shown in Table 2&Fig. 2. The differences between the three groups were statistically significant from the fourth week onwards (P<0.05). The initial number of white blood cells for all groups was slightly similar. However, over the six weeks, Group B (fed with an 85% carbohydrate diet) had consistently higher numbers of white blood cells than Group A (fed with a normal diet) and Group C (fed with a diet containing 65% protein). At the end of the experiment's sixth week, the number of white blood cells in rabbits was significantly higher (23.060±1.0126 million per microliter) in Group B than in Group A (9.742±0.29

million per microliter) and Group C (8.60±0.45 million per microliter).

Table-2 Effect of different diets on the white blood cell (WBC) count

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
WBC	1^{st}	9.82±0.36	10.157±0.5029	7.63±0.49	0.741
	2^{nd}	10.18±0.45	12.610±0.4347	6.98±0.30	0.651
	3^{rd}	9.05±0.43	15.603±1.1946	7.75±0.57	0.077
	4^{th}	10.79±0.72	19.452±1.1814	8.09±0.28	0.031
	5^{th}	10.68±0.67	21.473±1.1975	7.62±0.31	0.029
	6^{th}	9.742±0.29	23.060±1.0126	8.60±0.45	0.029

Fig= 2 Effect of different diets on the white blood cell (WBC) count



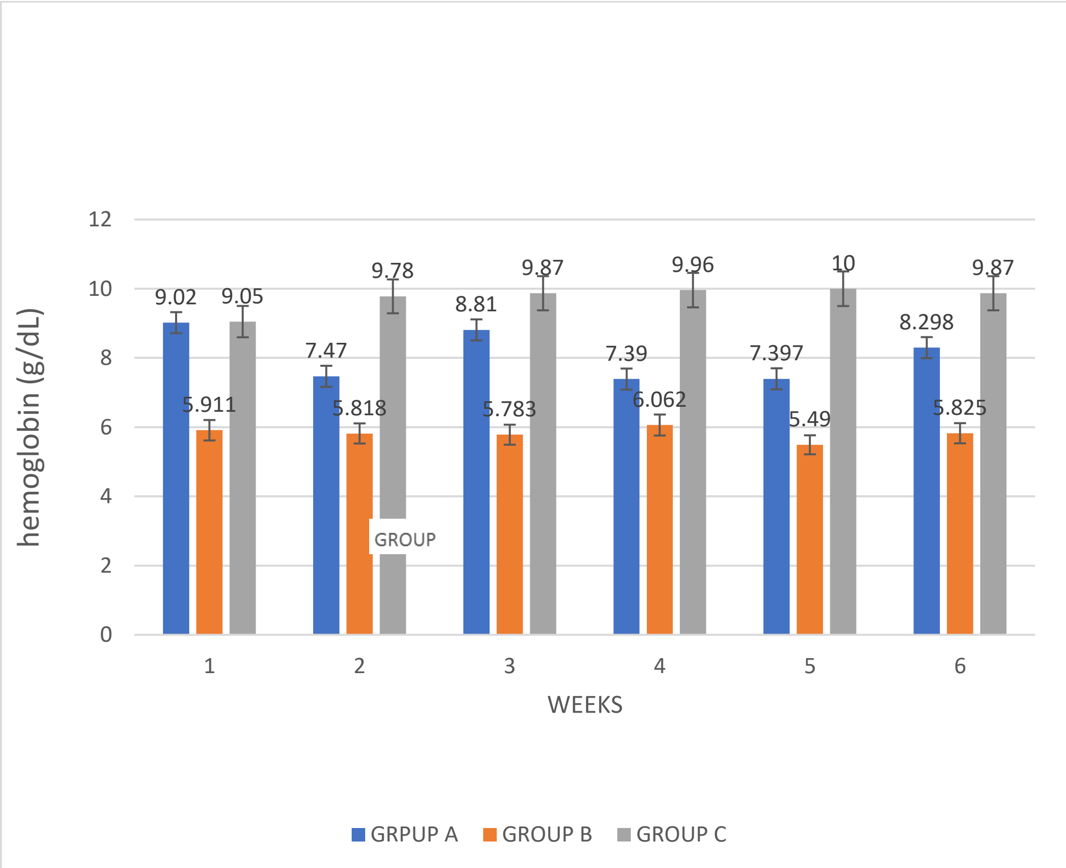
4.3 The impact of various diets on the blood haemoglobin level in rabbits

The table-3 & Fig. 3 compare the blood haemoglobin levels in rabbits over six weeks under different dietary conditions. Group A (received a basal diet), Group B (received an 85% high-carbohydrate diet), and Group C (received a 65% high-protein diet). The initial haemoglobin levels for all groups were not similar. Group C had the highest haemoglobin level, followed by Group A and Group B. Over the six weeks, Group C showed a significant increase in haemoglobin level, while Group A and Group B showed slight fluctuations. At the end of the experiment (sixth week), the haemoglobin level of the rabbits was significantly higher (9.8733±0.3715 g/dL) in Group C than in Group A (8.2983±0.9432 g/dL) and Group B (5.8250±0.0810 g/dL). The difference between Group C and the other groups was statistically significant in the first, second, fifth, and sixth weeks (P<0.05). The findings of these results suggested that a high-protein diet may increase the haemoglobin level in rabbits.

Table -3The impact of various diets on the blood haemoglobin level in rabbits

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
Haemoglobin	1st	9.02± 0.360	5.911± 0.16	9.05± 0.033	0.0318
	2nd	7.47± 0.531	5.818± 0.13	9.78± 0.356	0.0278
	3rd	8.81± 0.582	5.783± 0.17	9.87± 0.242	0.2610
	4th	7.39±0.546	6.062± 0.206	9.96± 0.434	0.1366
	5th	7.397±0.292	5.490± 0.035	10.00± 0.178	0.0013
	6th	8.298± 0.943	5.825± 0.081	9.87 3± 0.37	0.0042

Fig- 3 The impact of various diets on the blood haemoglobin level in rabbits



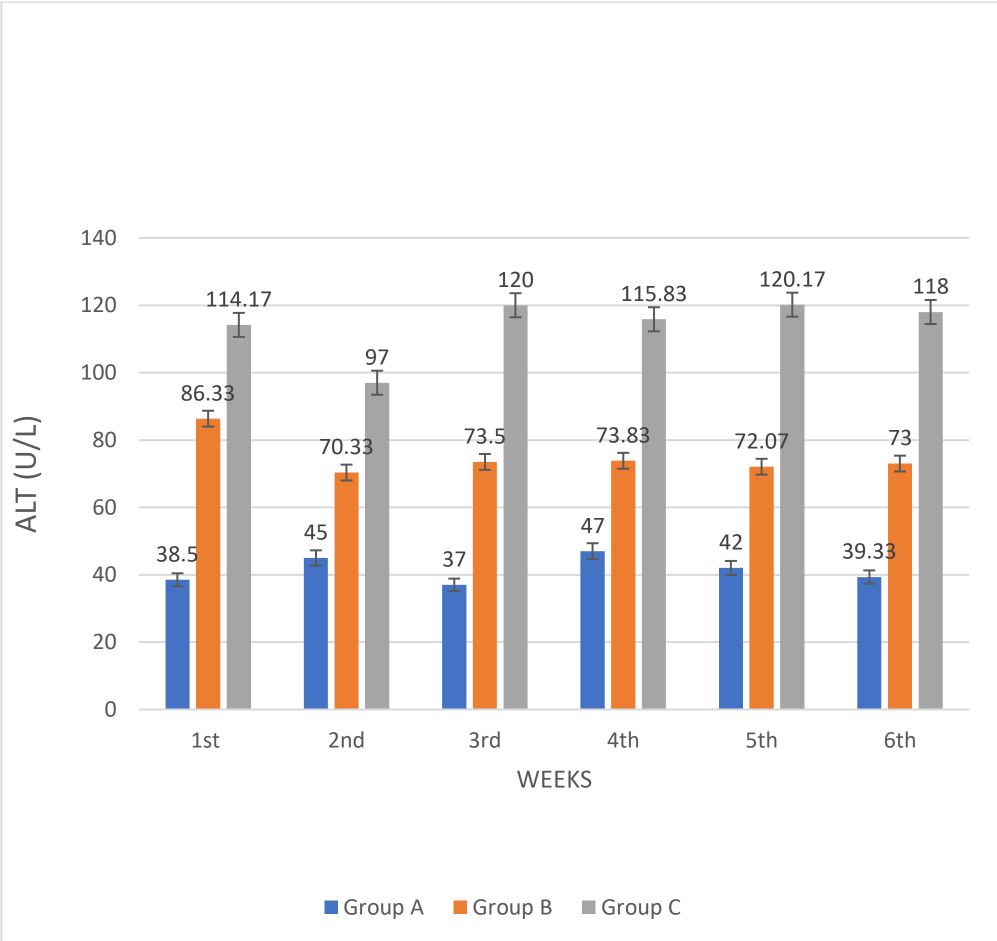
4 Effect of different diets on the blood alanine aminotransferase (ALT) level in rabbits

The effect of different diets on the alanine aminotransferase (ALT) level in rabbits over six weeks was investigated and presented in Table-4 & Fig-4. The differences between the three groups were statistically significant except in the fourth week (P<0.05). The initial levels of ALT for all groups were not similar. However, over the six weeks, Group C (fed with a diet containing 65% protein) had consistently higher levels of ALT than Group A (fed with a normal diet) and Group B (fed with an 85% carbohydrate). At the end of the experiment (sixth week), the level of ALT of rabbits was slightly higher (118.00±4.27 units per litre) in Group C than in Group A (39.33±3.32 units per litre) and Group B (73.00±4.48 units per litre), but the difference was significant. This suggests that a high protein-rich diet had a significant effect on the liver function of the rabbits.

Table-4 Effect of different diets on the blood alanine aminotransferase (ALT) level

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
Alanine aminotransferase (ALT)	1st	38.50±1.38	86.33±2.62	114.17±4.39	0.038
	2nd	45.00±1.57	70.33±4.95	97.00±5.41	0.048
	3rd	37.00±1.59	73.50±4.42	120.00±3.81	0.018
	4th	47.00±2.39	73.83±3.78	115.83±6.39	0.122
	5th	42.00±3.09	72.07±5.86	120.17±3.18	0.044
	6th	39.33±3.32	73.00±4.48	118.00±4.27	0.031

Fig-4 Effect of different diets on the blood alanine aminotransferase (ALT) level in rabbits



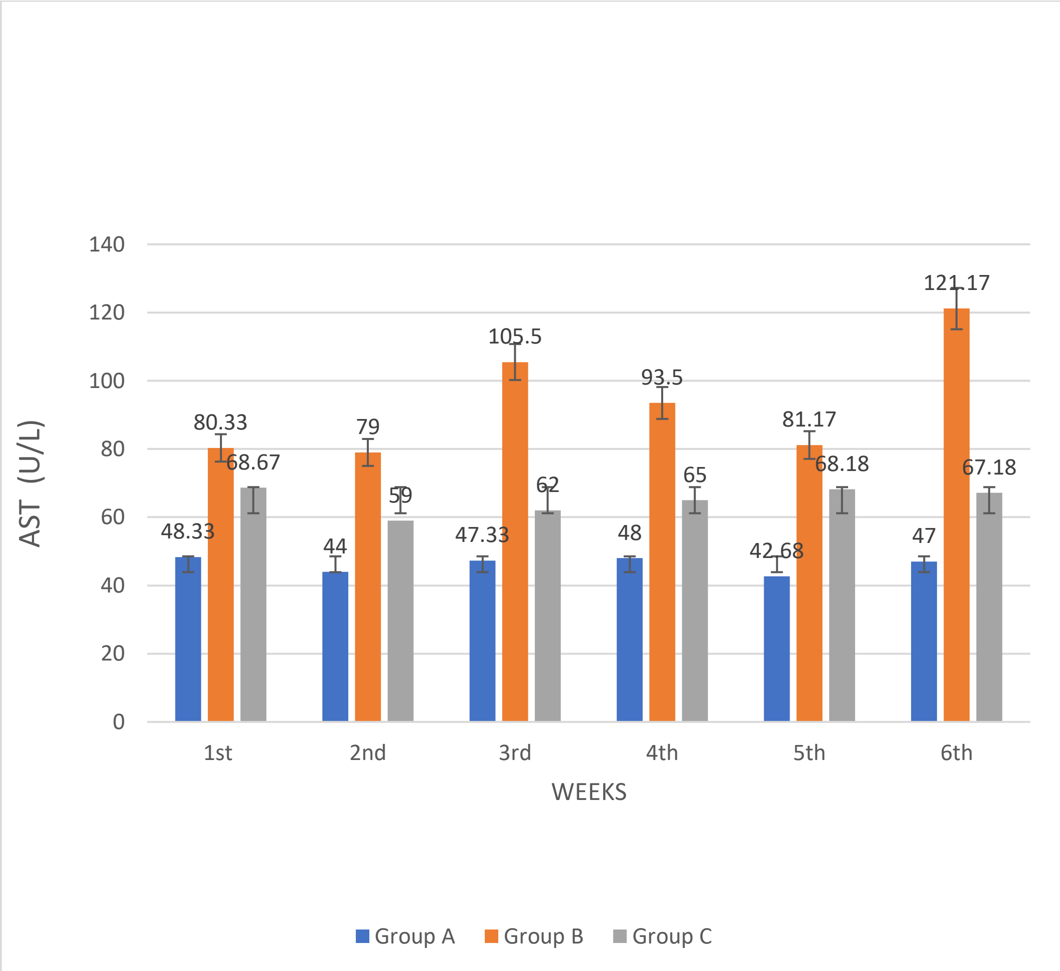
5. Effect of different diets on the blood aspartate aminotransferase (AST) level

The effect of different diets on the aspartate aminotransferase (AST) level in rabbits over six weeks was investigated and is presented in Table 5 & Fig. 5. The differences between the three groups were statistically significant except for the first and sixth weeks ($P<0.05$). The initial AST levels of all groups were not similar. However, over the six weeks, Group B had consistently higher AST levels than Group A and Group C. At the end of the experiment (sixth week), the AST level of the rabbits was significantly higher (121.17±5.49 units per litre) in Group B than in Group A (47.00±2.21 units per litre) and Group C (67.18±3.52 units per litre). The data showed that the high-carbohydrate diet impaired the rabbits' liver function of the rabbits.

Table-5 Effect of different diets on the blood aspartate aminotransferase (AST) level

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
Aspartate amino transferase (AST)	1st	48.33±4.21	80.33±3.051	68.67±4.36	0.723
	2nd	44.00±2.16	79.00±3.23	59.00±7.47	0.027
	3rd	47.33±2.36	105.50±5.89	62.00±5.58	0.047
	4th	52.00±1.73	93.50±2.49	65.00±5.98	0.024
	5th	42.68±2.40	81.17±5.08	68.18±6.15	0.016
	6th	47.00±2.21	121.17±5.49	67.18±3.52	0.168

Fig-5 Effect of different diets on the blood aspartate aminotransferase (AST) level



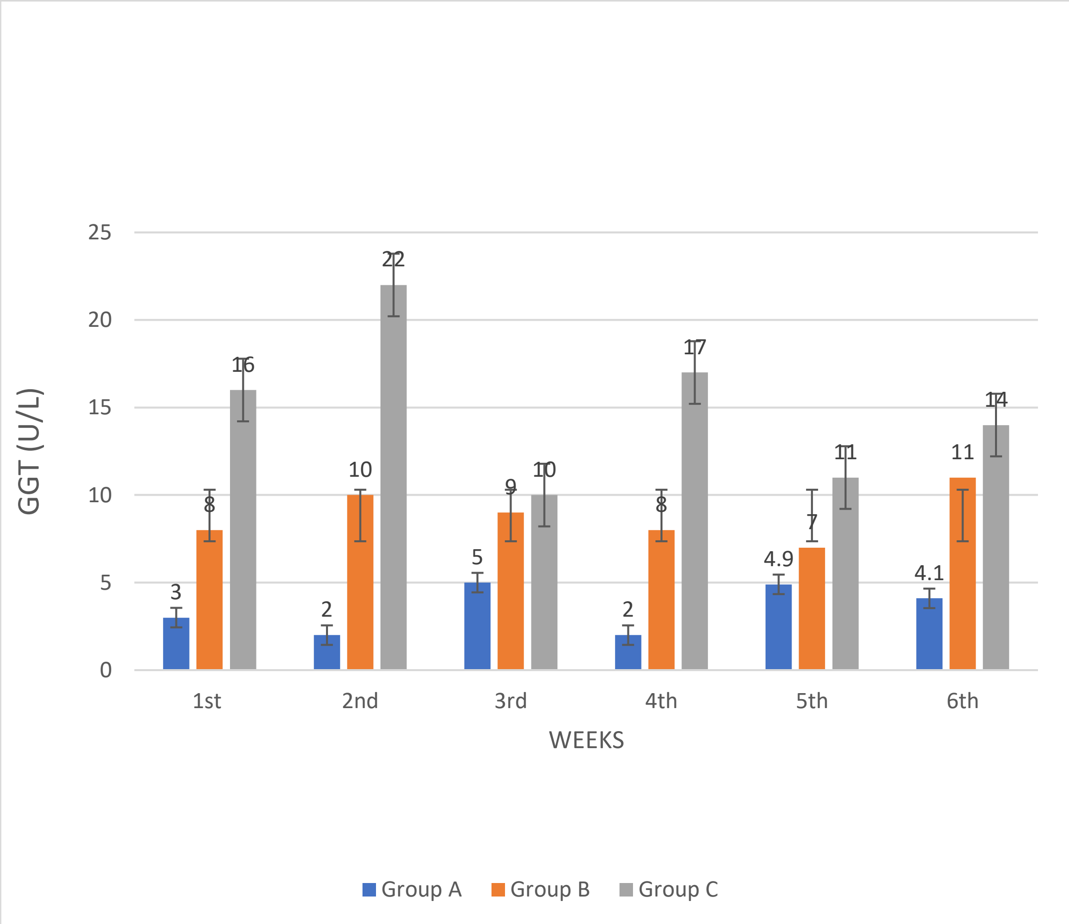
6. Effect of different diets on the blood gamma-glutamyl transferase (GGT) level

The impact of various diets on the gamma-glutamyl transferase (GGT) levels in rabbits over six weeks is depicted in Table 6 & Fig. 6. The differences between the three groups were statistically significant in the second, fourth, and sixth weeks (P<0.05). The initial GGT levels for all groups were not similar. However, over the six weeks, Group C (fed with a diet containing 65% protein) had consistently higher GGT levels than Group A (fed with a normal diet) and Group B (fed with an 85% carbohydrate diet). At the end of the experiment (sixth week), the level of GGT of rabbits was significantly higher (14.00±1.46 units per litre) in Group C than in Group A (4.10±0.27 units per litre) and Group B (11.00±0.97 units per litre).

Table -6 Effect of different diets on the blood gamma-glutamyl transferase (GGT) level

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
Glutamyl-transferase (GGT)	1st	3.00±0.49	8.00±0.52	16.00±1.095	0.1407
	2nd	1.00±0.19	10.00±0.86	22.00±1.37	0.0027
	3rd	5.00±0.45	9.00±0.45	10.00±1.00	0.1167
	4th	2.00±0.26	8.00±0.52	17.00±2.29	0.0001
	5th	6.02±0.34	7.00±0.58	11.00±1.03	0.0739
	6th	4.10±0.27	11.00±0.97	14.00±1.46	0.0097

Fig-6 Effect of different diets on the blood gamma-glutamyl transferase (GGT) level



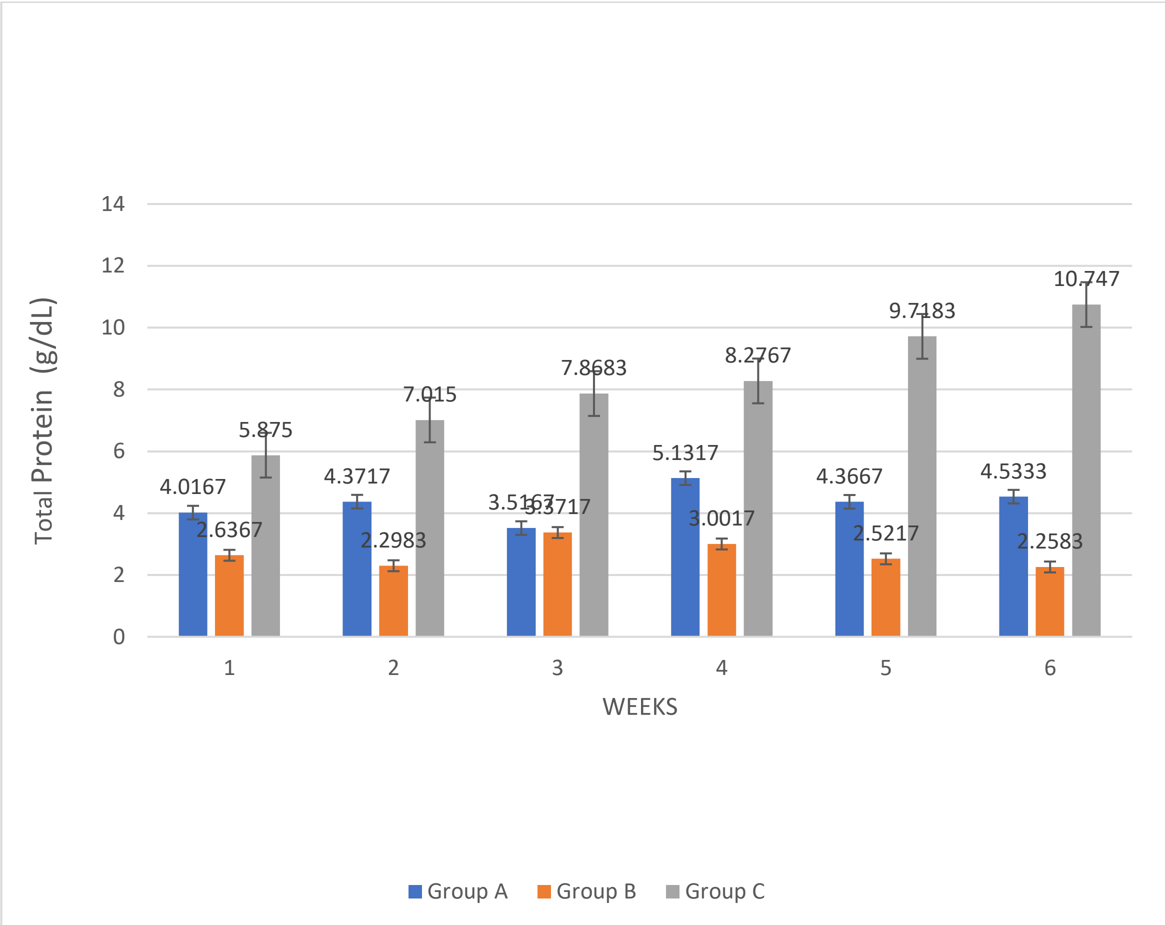
7. The impact of various diets on the blood total protein levels in rabbits

The impact of various diets on the total protein levels in rabbits over six weeks is presented in Table-7 & Fig-7. The differences between the three groups were statistically significant in all weeks except the first week (P<0.05). The initial total protein levels for all groups were not similar. However, over the six weeks, Group C (fed with 65% protein) consistently had higher total protein levels than Group A (fed with a normal diet) and Group B (fed with a diet containing 85% carbohydrate). At the end of the experiment (sixth week), the total protein level of the rabbits was significantly higher (10.747±0.2028 g/dL) in Group C than in Group A (4.5333±0.4232 g/dL) and Group B (2.2583±0.8629 g/dL). This suggests that a high-protein-rich diet increases the total protein level in rabbits.

Table-7 The impact of various diets on the blood total protein levels in rabbits

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
Total Protein	1st	4.0167±0.36	2.6367±0.219	5.8750 ±0.22	0.431
	2nd	4.3717±0.45	2.2983±0.277	7.0150 ±0.13	0.046
	3rd	3.5167±0.45	3.3717±0.901	7.8683±0.211	0.016
	4th	5.1317±0.37	3.0017±0.911	8.2767 ±0.30	0.038
	5th	4.3667±0.47	2.5217±0.853	9.7183 ±0.14	0.004
	6th	4.5333±0.42	2.2583±0.863	10.747 ±0.20	0.016

Fig-7 The impact of various diets on Total protein levels



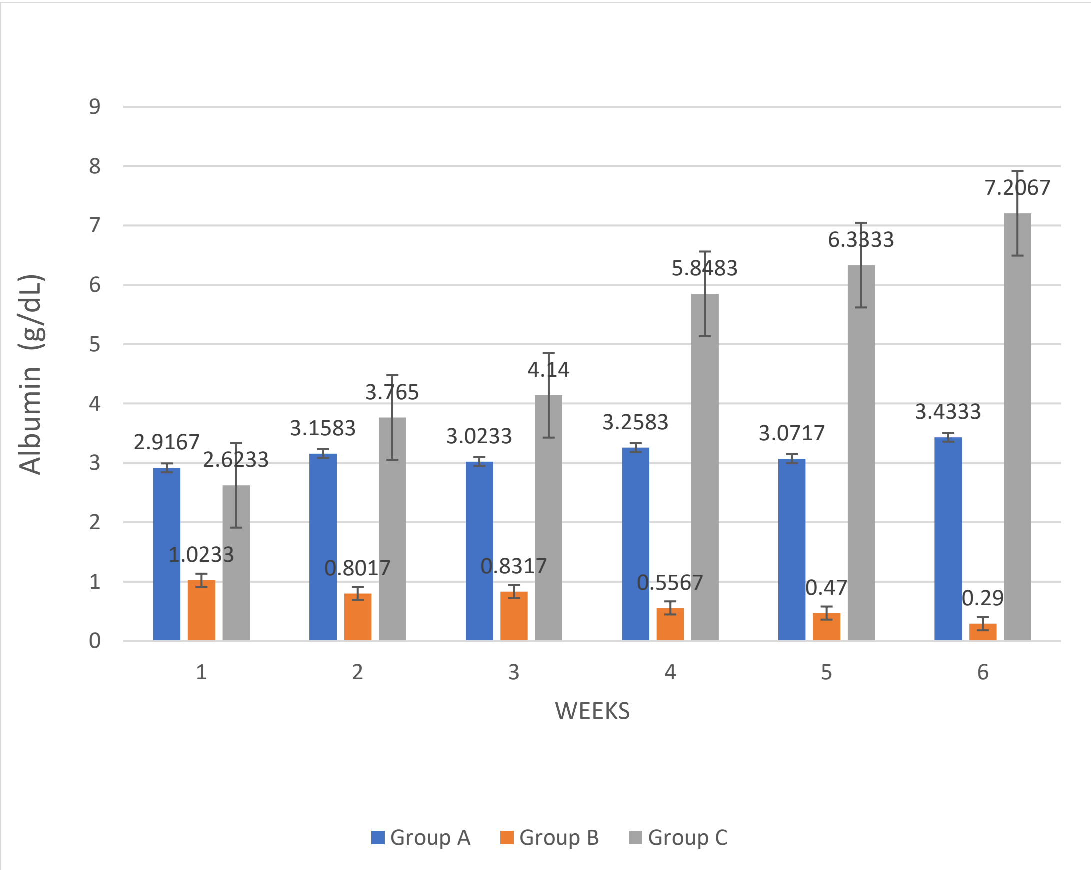
8. The impact of various diets on blood albumin levels

The effect of different diets on the albumin levels in rabbits over six weeks is shown in Table-8 & Fig-8. The differences between the three groups were statistically significant in all weeks (P<0.05). The initial albumin levels for all groups were similar. However, over the six weeks, Group C (fed with a diet containing 65% protein) had consistently higher levels of albumin than Group A (fed with a normal diet) and Group C (fed with an 85% carbohydrate). At the end of the experiment (sixth week), the albumin level of rabbits was significantly higher (7.2067±0.2557 g/dL) in Group C than in Group A (3.4333±0.2578 g/dL) and Group B (0.2900±0.0347 g/dL). The observed data suggest that a high-protein diet may increase the albumin level in rabbits.

Table-8 The impact of various diets on blood Albumin levels

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
Albumin	1st	2.9167±0.1778	1.0233±0.1365	2.6233±0.1613	0.046
	2nd	3.1583±0.2653	0.8017±0.1252	3.7650±0.2361	0.027
	3rd	3.0233±0.2340	0.8317±0.1019	4.1400±0.1295	0.017
	4th	3.2583±0.2216	0.5567±0.0720	5.8483±0.3134	0.022
	5th	3.0717±0.2992	0.4700±0.0336	6.3333±0.2369	0.001
	6th	3.4333±0.2578	0.2900±0.0347	7.2067±0.2557	0.002

Fig-8 The impact of various diets on blood Albumin levels



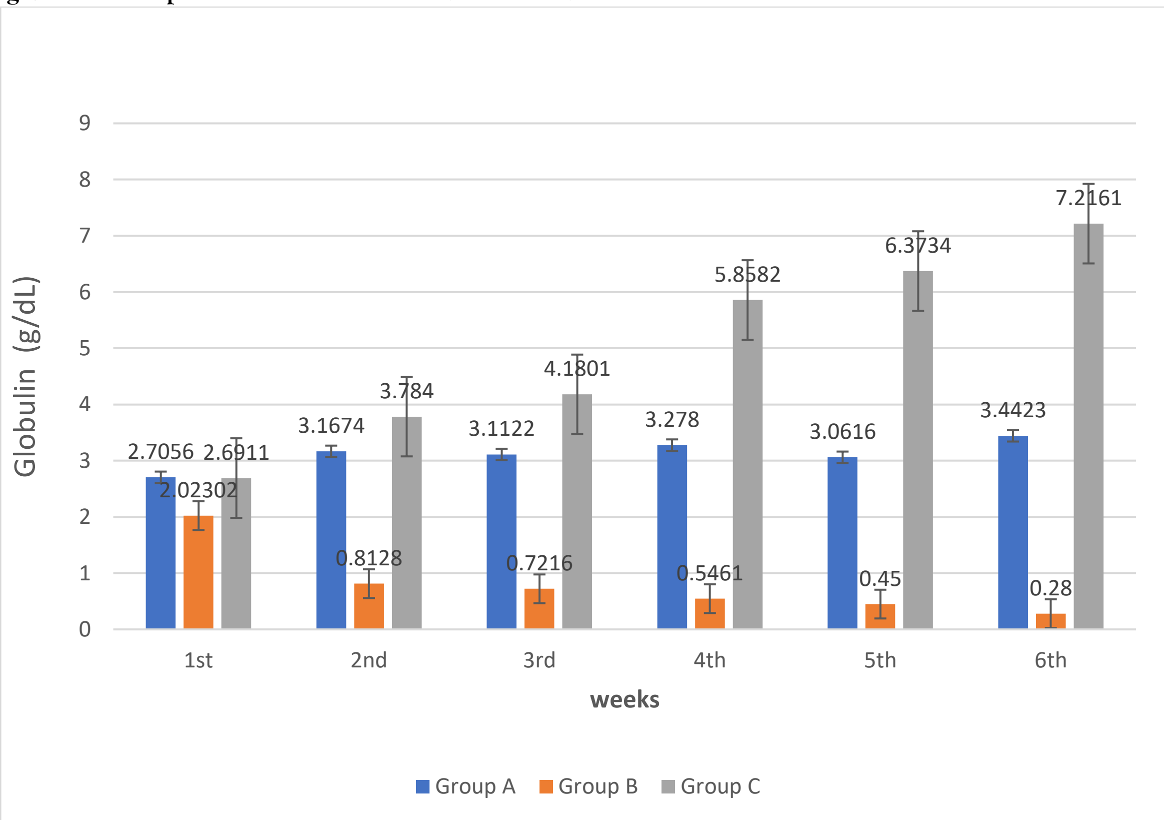
9. The impact of various diets on blood Globulin levels

The effect of different diets on the globulin level in rabbits over six weeks is shown in Table-9 & Fig-9. The initial globulin5 levels for all groups were similar. However, over the six weeks, Group C (7.2161±0.2659) differed from Group A (3.4423±0.2776 g/dL) and Group B (0.2800±0.0345 g/dL). The table clarifies the impact of different dietary regimens on the globulin levels in rabbits over a six-week period. Statistical analysis reveals significant differences among the three groups throughout the study duration (P < 0.05). Initially, baseline globulin levels were similar across all groups. However, by the conclusion of the six-week period, notable discrepancies in globulin levels emerged. Group C exhibited markedly higher globulin levels (7.2161±0.2659 g/dL) compared to Group A (3.4423±0.2776 g/dL) and Group B (0.2800±0.0345 g/dL). These findings underscore a distinct relationship between dietary protein intake and globulin levels in rabbits, suggesting that a diet with increased protein content may lead to elevated globulin levels over time.

Table-9 The impact of various diets on blood globulin levels

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
Globulin	1st	2.7056±0.0.0875	2.02302±0.1254	2.6911±0.1510	0.043
	2nd	3.1674±0.28657	0.8128±0.1251	3.7840±0.2251	0.024
	3rd	3.1122±0.2230	0.7216±0.1017	4.1801±0.1194	0.015
	4th	3.2780±0.2519	0.5461±0.0621	5.8582±0.3133	0.020
	5th	3.0616±0.2893	0.4500±0.0338	6.3734±0.2268	0.003
	6th	3.4423±0.2776	0.2800±0.0345	7.2161±0.2659	0.001

Fig- 9 The impact of various diets on blood Globulin levels



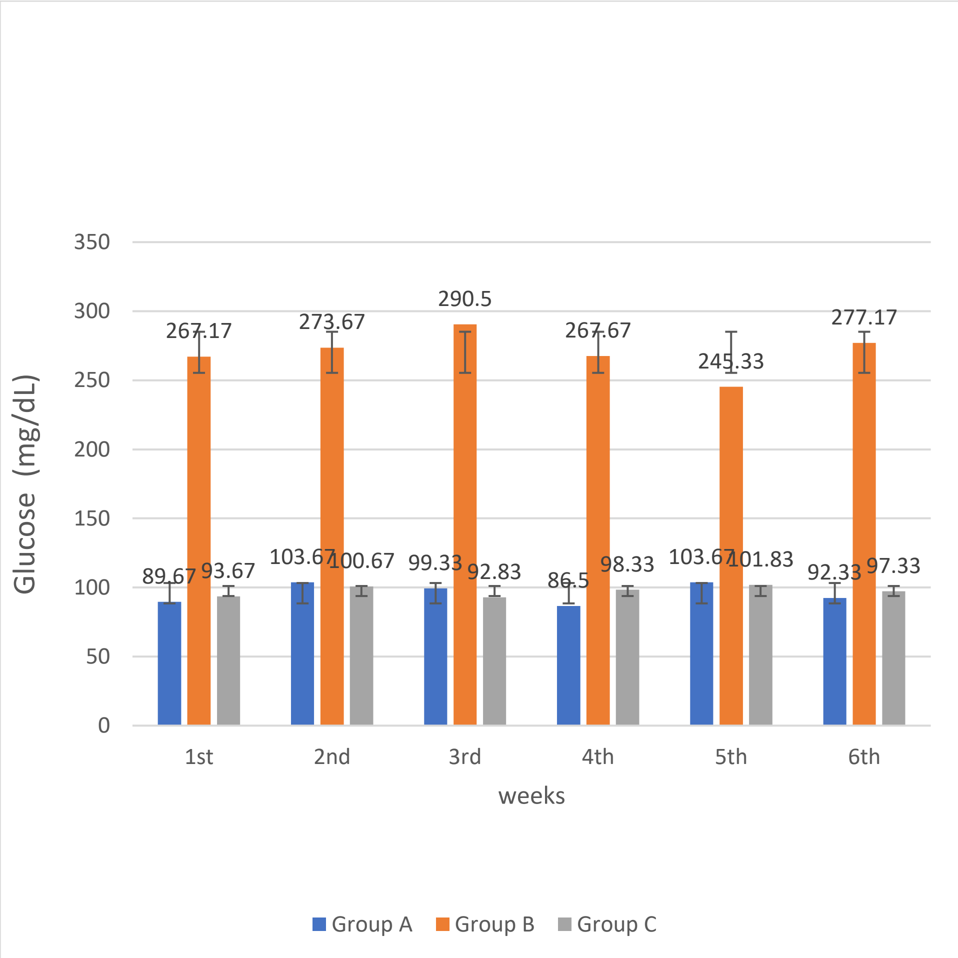
10. Effect of different diets on the blood Glucose level

The effect of different diets on the blood glucose level in rabbits over six weeks was investigated and presented in Table 19 & Fig. 10. The differences between the three groups were statistically significant in all weeks except the second and third weeks (P<0.05). The initial glucose levels for all groups were not similar. However, over the six weeks, Group B (fed with an 85% carbohydrate diet) had consistently higher glucose levels than Group A (fed with a normal diet) and Group C (fed with a diet containing 65% protein enriched with vitamins and minerals). At the end of the experiment (sixth week), the glucose level of rabbits was significantly higher (277.17 ± 20.511 mg/dL) in Group B than in Group A (92.333 ± 3.4801 mg/dL) and Group C (10.33 ± 8.597 mg/dL). This suggests that a high-carbohydrate diet may increase the glucose level in rabbits.

Table-10 **Effect of Different Diets on the Blood glucose level**

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
Glucose	1st	89.67± 6.78	267.17 ± 17.92	93.67± 5.81	0.0177
	2nd	103.67 ± 6.78	273.67± 13.10	100.67± 4.86	0.0945
	3rd	99.33± 9.99	290.50± 12.62	92.83 ± 4.09	0.0855
	4th	86.50 ± 2.56	267.67± 5.91	98.33 ± 1.41	0.0127
	5th	103.67± 10.06	245.33± 7.44	101.83 ± 2.53	0.0317
	6th	92.33 ± 3.48	277.17 ± 20.51	10.33 ± 8.59	0.0029

Fig-10 **Effect of different diets on the Blood glucose level**



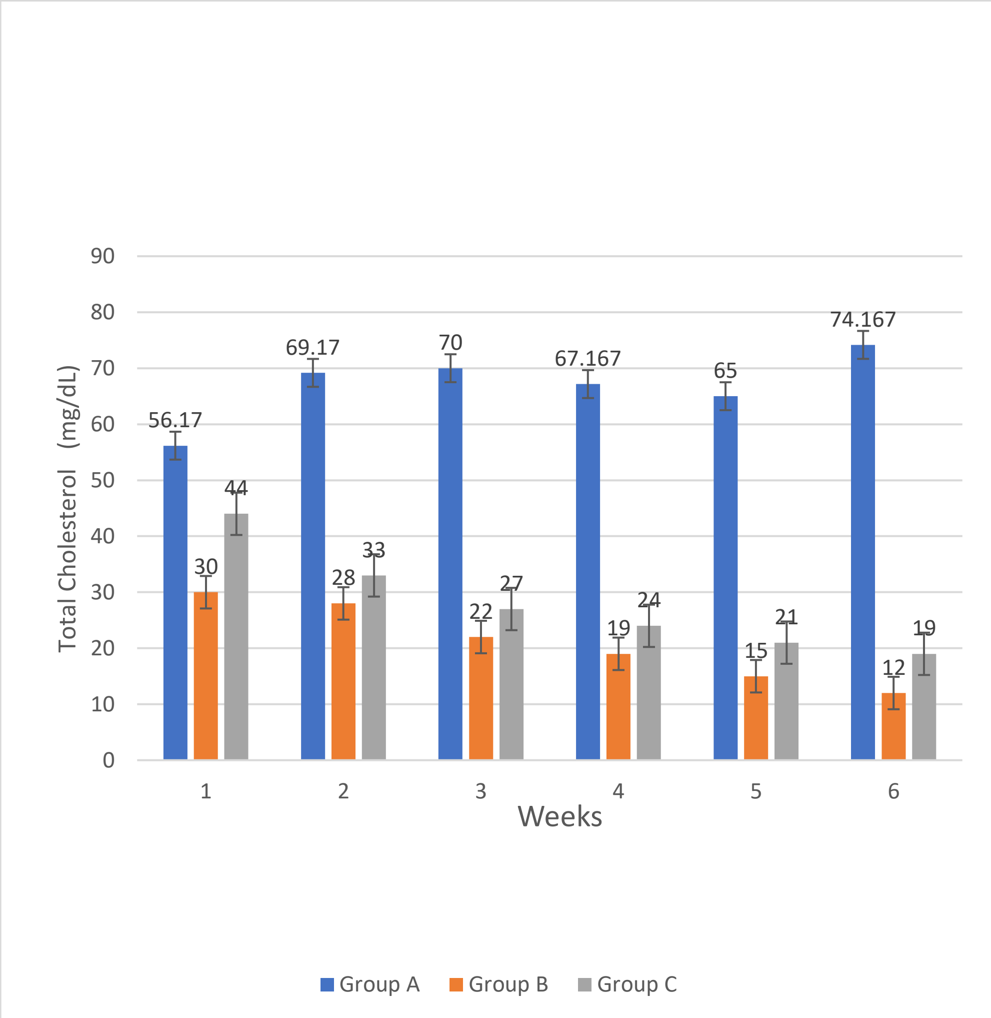
11. The impact of various diets on the blood total cholesterol levels in rabbits

The effect of different diets on the total cholesterol level in rabbits over six weeks is shown in Table 11 &Fig. 11. The differences between the three groups were statistically significant only in the fifth week ($P<0.05$). The initial total cholesterol levels for all groups were not similar. However, over the six weeks, Group A (fed with a normal diet) had consistently higher levels of total cholesterol than Group C (fed with 65% protein) and Group B 85% Carbohydrate. At the end of the experiment (sixth week), the total cholesterol level of rabbits was higher (74.167±2.7739 mg/dL) in Group A than in Group B (12.00±1.18 mg/dL) and Group C (19.00±1.59 mg/dL), but the difference was not significant. This suggests that a normal diet may increase the total cholesterol level in rabbits.

Table -11 The impact of various diets on the blood total cholesterol levels in rabbits

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
Total Cholesterol	1st	56.17±2.77	30.00±1.86	44.00±1.93	0.6230
	2nd	69.17±2.14	28.00±1.37	33.00±1.59	0.6152
	3rd	70.000±2.62	22.00±0.93	27.00±1.89	0.1202
	4th	67.167±1.51	19.00±1.24	24.00±1.59	0.8568
	5th	65.000±2.83	15.00±1.34	21.00±0.77	0.0270
	6th	74.167±2.77	12.00±1.18	19.00±1.59	0.1755

Fig-11 The impact of various diets on the blood Total Cholesterol levels in rabbits



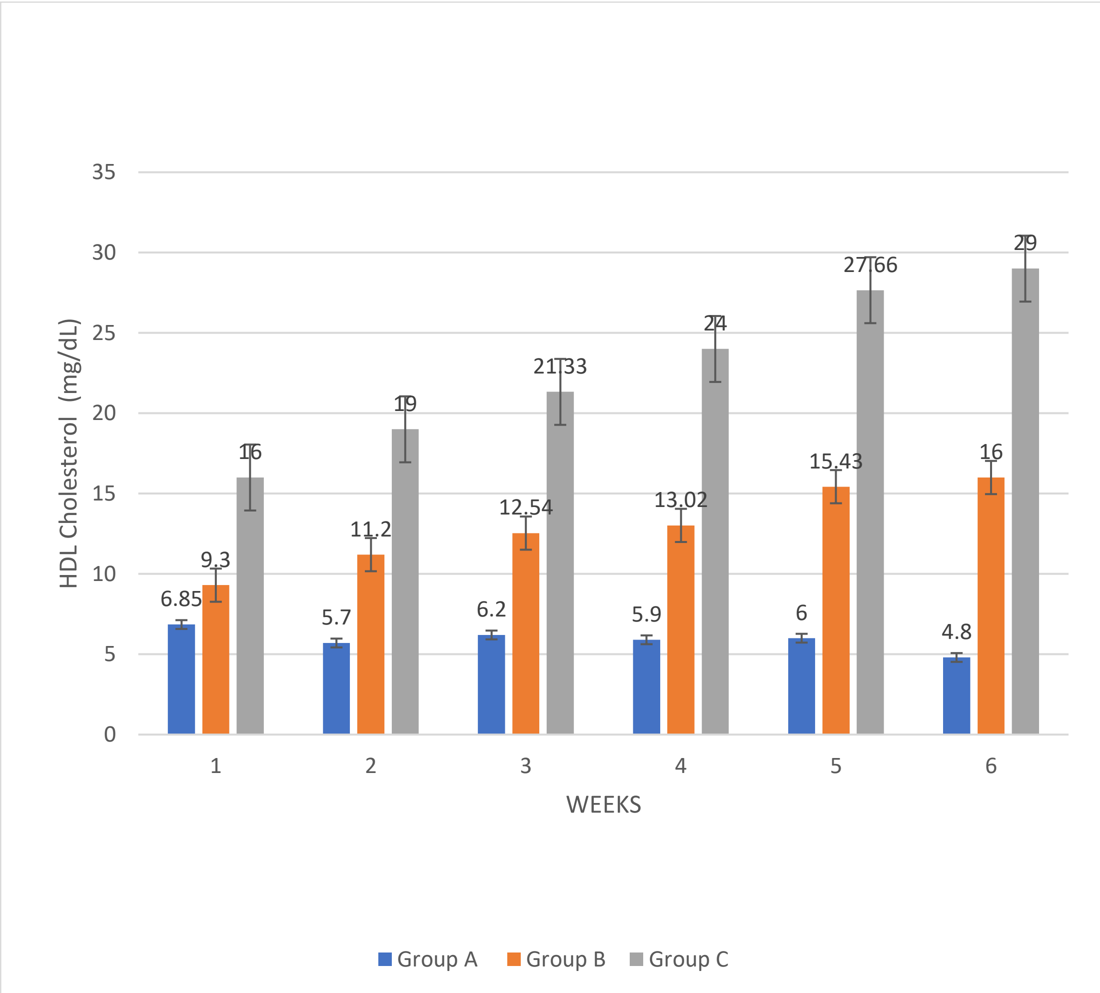
12. Effect of different diets on the blood HDL cholesterol level

The table illustrates the impact of various diets on the blood HDL cholesterol level in rabbits over six weeks. The differences between the three groups were statistically significant in all weeks ($P<0.05$). The initial HDL cholesterol levels for all groups were not similar. However, over the six weeks, Group C had consistently higher levels of HDL cholesterol than Group A and B. At the end of the experiment (sixth week), the HDL cholesterol level of rabbits was significantly higher (29.00±1.32 mg/dL) in Group C than in Group A (4.80±0.14 mg/dL) and Group B (16.00±1.65 mg/dL). The findings of these results showed that a high-protein diet may increase the HDL cholesterol level in rabbits.

Table-12 Effect of different diets on the blood HDL cholesterol level

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
HDL cholesterol	1st	6.85±0.20	9.30±0.35	16.00±1.48	0.0002
	2nd	5.70±0.15	11.20±0.71	19.00±1.18	0.0018
	3rd	6.20±0.09	12.54±0.62	21.33±1.28	0.0001
	4th	5.90±0.26	13.02±0.81	24.00±0.86	0.0042
	5th	6.00±0.57	15.43±0.74	27.66±2.23	0.0083
	6th	4.80±0.14	16.00±1.65	29.00±1.32	0.0003

Fig-12 Effect of different diets on the blood HDL Cholesterol level



13. Effect of different diets on the blood LDL cholesterol level

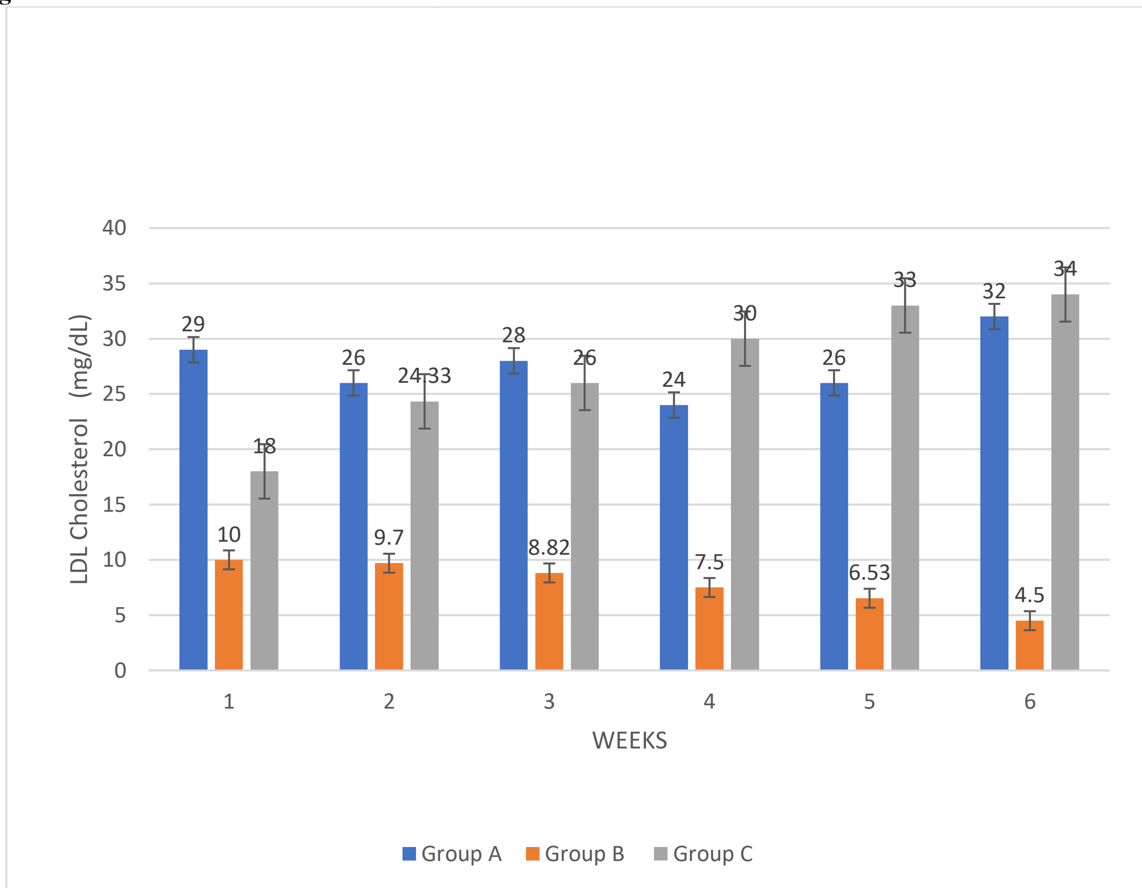
The table-13 & Fig-13 show the effect of different diets on the blood LDL cholesterol level in rabbits over six weeks. The initial LDL cholesterol levels for all groups were not similar. Group A (fed with a normal diet) had a higher LDL cholesterol level, followed by

Group C (fed with 65% protein) and Group B (85% carbohydrate). Over the six weeks, Group A and Group C showed similar trends of increasing LDL cholesterol levels, while Group B showed a decreasing trend. At the end of the experiment (sixth week), the LDL cholesterol level of rabbits was significantly lower (4.50±0.84 mg/dL) in Group B than in Group A (32.00±0.97 mg/dL) and Group C (34.00±3.12 mg/dL). The differences between the three groups were statistically significant in the second, fourth, fifth, and sixth weeks (P<0.05). The findings of these results indicated that a high-carbohydrate diet may decrease the LDL cholesterol level in rabbits.

Table-13 Effect of different diets on the blood LDL cholesterol level

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
LDL Cholesterol	1st	29.00±0.97	10.00±1.67	18.00±2.37	0.1908
	2nd	26.00±1.32	9.70±0.89	24.33±2.60	0.0495
	3rd	28.00±2.14	8.82±0.86	26.00±1.93	0.1659
	4th	24.00±1.95	7.50±0.27	30.00±1.98	0.0018
	5th	26.00±2.02	6.53±0.84	33.00±2.98	0.0013
	6th	32.00±0.97	4.50±0.84	34.00±3.12	0.0057

Fig-13 Effect of different diets on the blood LDL cholesterol level



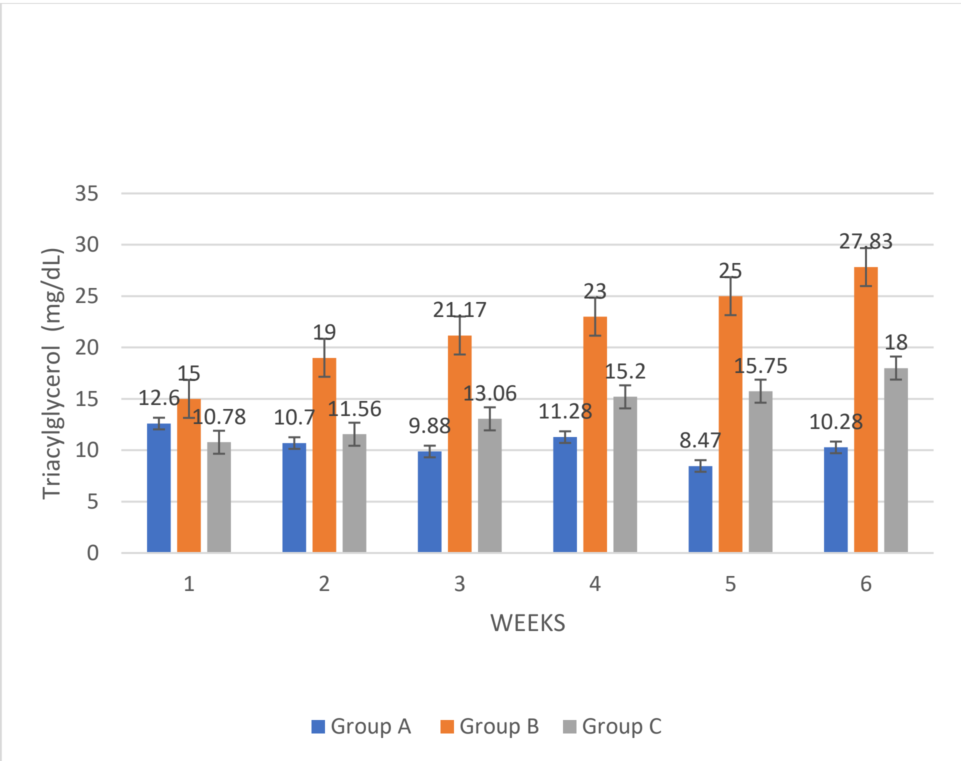
14. Effect of different diets on the blood triacylglycerol level

The table-14 & Fig-14 compares the blood triacylglycerol levels in rabbits over six weeks under different dietary conditions. Group A (received a basal diet), Group B (received an 85% high-carbohydrate diet), and Group C (received a 65% high-protein diet). The initial triacylglycerol levels for all groups were not similar. Group B had the highest triacylglycerol level, followed by Groups A and C. Over the six weeks, Group B showed a sharp increase in triacylglycerol levels, while Group A and C showed moderate increases. At the end of the experiment (sixth week), the triacylglycerol level of rabbits was significantly higher (27.83±3.10 mg/dL) in Group B than in Group A (10.28±0.66 mg/dL) and Group C (18.00±0.96 mg/dL). The differences between the three groups were statistically significant in the third and sixth weeks ($P<0.05$). The findings of these results suggested that a high-carbohydrate diet may raise the triacylglycerol level in rabbits.

Table-14 Effect of different diets on the blood triacylglycerol level

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
Triacylglycerol	1st	12.60±1.20	15.00±1.59	10.78±1.19	0.7751
	2nd	10.70±0.89	19.00±1.82	11.56±0.98	0.2286
	3rd	9.88±0.92	21.17±2.77	13.06±0.76	0.0102
	4th	11.28±0.91	23.00±0.93	15.20±1.60	0.3682
	5th	8.47±0.97	25.00±2.03	15.75±1.01	0.1817
	6th	10.28±0.66	27.83±3.10	10.28±0.66	0.0030

Fig-14 Effect of different diets on the blood triacylglycerol level



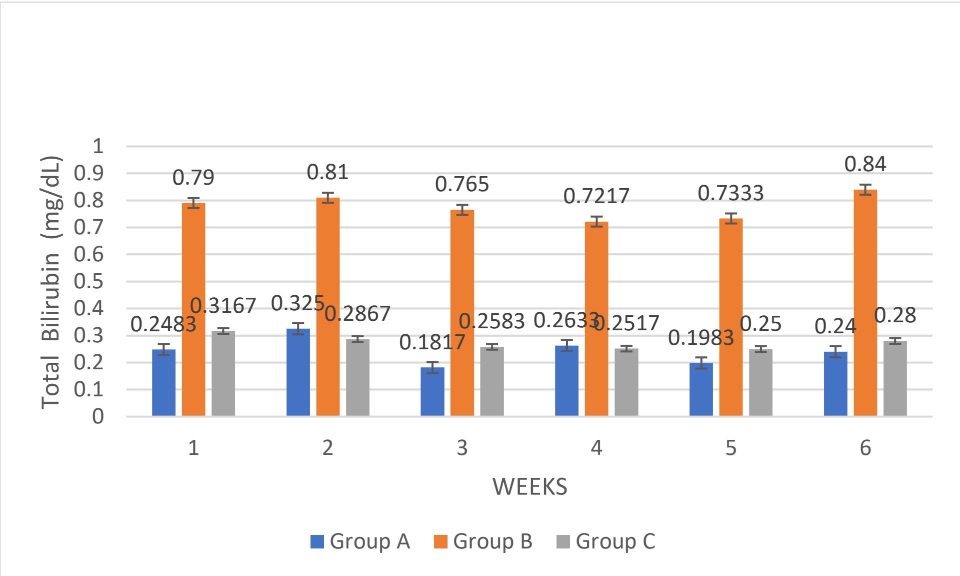
15. Effect of different diets on the blood bilirubin level

The Table-15 &Fig-15 compare the blood total bilirubin levels in rabbits over six weeks under different dietary conditions. Group A (received a basal diet), Group B (received an 85% high-carbohydrate diet), and Group C (received a 65% high-protein diet). The initial total bilirubin levels for all groups were not similar. Group B had the highest total bilirubin level, followed by Group C and Group A. Over the six weeks, Group B maintained a high total bilirubin level, while Group A and Group C fluctuated slightly. At the end of the experiment (sixth week), the total bilirubin level of rabbits was significantly higher (0.8400±0.0603 mg/dL) in Group B than in Group A (0.2400±0.0390 mg/dL) and Group C (0.2800±0.0493 mg/dL). The difference between Group B and the other groups was statistically significant in the first week (P<0.05). The findings of these results suggested that a high-carbohydrate diet may increase the total bilirubin level in rabbits.

15. Effect of different diets on the blood bilirubin level

Parameter	Weeks	Group A Mean±SE	Group B Mean±SE	Group C Mean±SE	P value
Total Bilirubin	1st	0.2483±0.0488	0.7900±0.1125	0.3167±0.0279	0.0149
	2nd	0.3250±0.1203	0.8100±0.1001	0.28670.0476	0.1707
	3rd	0.1817±0.0187	0.7650±0.0985	0.2583±0.0119	0.1110
	4th	0.2633±0.0493	0.7217±0.0500	0.2517±0.0334	0.6483
	5th	0.1983±0.0204	0.7333±0.0643	0.2500±0.0392	0.0410
	6th	0.8400±0.0603	0.8400±0.0603	0.2800±0.0493	0.5624

Fig-15 Effect of different diets on the blood Total Bilirubin level



DISCUSSIONS

This study was done on three groups, as are the following, group A, which was fed a basal diet; group B, which was treated with a high-carbohydrate diet; and group C, which was treated with a high-protein diet. Rabbits, the herbivorous creatures, survive on a diet that primarily consists of grasses and herbs. Their diet is rich in fibre, which they ferment in the hindgut to produce cecotrophs. These cecotrophs are then reingested to obtain essential nutrients (Meredith, 2015). Rabbits require a balanced and complete diet that includes an adequate amount of energy, protein, fibre, vitamins, minerals, and water to ensure their health and well-being. This study aims to examine the effect of a high protein and high carbohydrate diet on different physiological factors in rabbits. The study intends to assess the possible consequences of these diets on the liver's function, glucose levels, and haematological responses in rabbits.

In the study, it was observed that rabbits in group C (10.98±0.36 million per microliter) consistently showed higher values of red blood cells than those in group A (7.03±0.46 million per microliter) and group B (5.037±0.33 million per microliter). These results are in line with previous research efforts (Varga, 2014; Varga, 2015).

The results indicate that group B (23.060±1.0126 million per microliter) had an impact on the white blood cell count, causing it to rise above the normal value when compared with group A (9.742±0.29 million per microliter) and group C (8.60±0.45 million per microliter). This finding is in agreement with the research conducted (Harcourt-Brown et al., 2020; MSD Veterinary Manual, (2021). On the other hand, studies indicate that high-carbohydrate diets may cause a reduction in haemoglobin, red blood cell count, and white blood cell count in rabbits (Volek et al., 2009; Bazzano et al., (2014). These studies suggested that diets high in carbohydrates may lead to anaemia and immunosuppression. These alterations may be a result of ageing, which can impact the composition and function of white blood cells. Younger animals may still be in the process of developing and maturing their immune systems, while older animals may experience changes associated with aging.

The study's findings indicate that the group receiving treatment (Group C) (9.8733±0.3715 g/dL) had higher levels of haemoglobin when compared with Group A (8.2983±0.9432 g/dL) and Group B (5.8250±0.0810 g/dL). Additionally, a study with similar parameters was previously conducted by (Lovati et al., 1990; Varga, 2014; Varga, 2015). However, there are also reports suggesting that high-protein diets can result in lower levels of red blood cells, haemoglobin, and white blood cells in rabbits. (Hosseini-Mansoub, & Bahrami, 2011) According to certain studies, rabbits may experience an increase in haemoglobin, red blood cell count, and white blood cell count when consuming high-carbohydrate diets. The studies suggest that diets high in carbohydrates may positively affect erythropoiesis, immune function, and inflammation. Conversely, some studies have also shown that rabbits may experience a decrease in haemoglobin, red blood cell count, and white blood cell count when consuming high-carbohydrate diets (Volek et al.; 2009; Bazzano et al., 2014). Similar studies were conducted in rats in which the high-protein diet elevated haemoglobin levels (Obikaonu, et al., 2012)

When an individual follows a diet that is high in carbohydrates or proteins, their body may require more fluids than usual. It's crucial to keep oneself adequately hydrated to prevent hemoconcentration, which can result in elevated haemoglobin levels in the body. A well-balanced diet that includes ample protein is crucial for the production of erythropoietin, a hormone that stimulates the growth of red blood cells. A diet that is rich in both carbohydrates and protein can play a significant role in supporting the body's overall nutritional needs, which in turn can promote erythropoiesis.

Based on the results, it was found that Group C (118.00±4.27 units per litre) had a greater impact on ALT levels than Group A (39.33±3.32 units per litre) and Group B (73.00±4.48 units per litre). The consumption of a high-protein diet was found to have a beneficial effect on the liver, leading to an increase in ALT levels. In previous studies, it was observed that when rabbits were given a high-protein diet, their ALT levels also increased (Oboh & Olumese, 2008).

According to the findings, it was observed that the impact of Group B (121.17±5.49 units per litre) on AST levels was significantly greater than that of Group A (47.00±2.21 units per litre) and Group C (67.18±3.52 units per litre). The liver experienced a boost in AST levels due to the consumption of a high-carbohydrate diet. Studies conducted on rabbits also supported this observation, as their AST levels increased when fed a high-carbohydrate diet (Oboh et al., 2007).

According to the findings, it was observed that the impact of Group C (14.00±1.46 units per litre) on GGT levels was more significant than that of Group A (4.10±0.27 units per litre) and Group B (11.00±0.97 units per litre). It was discovered that a high-protein diet had adverse effects on the liver, which caused an increase in GGT levels. Similar outcomes were observed in rats when they were given a high-protein diet, as their GGT levels also increased (Anyakudo, et al., 2017).

It has been discovered that Group C (10.747±0.2028 g/dL) had a more significant effect on the total protein levels when compared to Group A (4.5333±0.4232 g/dL) and Group B (2.2583±0.8629 g/dL). The research suggests that following a high-protein diet can hurt the blood, leading to an increase in total protein levels. Previous studies have also shown that individuals who were given a high-protein diet experienced an increase in their total protein levels (Oboh, et al., 2007).

The study results revealed that Group C (7.2067±0.2557 g/dL) had a more substantial effect on albumin levels compared to Group A (3.4333±0.2578 g/dL) and Group B (0.2900±0.0347 g/dL). It was observed that consuming a high-protein diet hurt the bloodstream, leading to a rise in albumin levels. Previous research also found similar outcomes wherein individuals who were given a high-protein diet experienced an increase in albumin levels (Oboh, et al., 2007).

According to the study, Group C had a more significant impact on globulin levels (7.2161±0.2659) compared to Group A (3.4423±0.2776) and Group B (0.2800±0.0345). The research indicated that consuming a high-protein diet can have adverse effects on blood, leading to an increase in globulin levels. Similar outcomes were observed in previous studies, wherein individuals who consumed a high-protein diet experienced a rise in globulin levels (Oboh, et al., 2008).

The findings indicate that Group B (277.17 ± 20.511 mg/dL) had a more significant effect on glucose levels than Group A (92.333 ± 3.4801 mg/dL) and Group C (10.33 ± 8.597 mg/dL). The study revealed that consuming a diet high in carbohydrates can adversely affect blood sugar levels, leading to an increase in glucose levels. These conclusions align with previous research that also found an increase in glucose levels among individuals who consumed a high-carbohydrate diet (Hilarious, & Johnson, 2012).

According to the findings, Group A (74.167±2.7739 mg/dL), which was on a normal diet, experienced an increase in total cholesterol levels compared to Group B (12.00±1.18 mg/dL) and Group C (19.00±1.59 mg/dL). Additionally, the high-carbohydrate diet hurt the total cholesterol level when given to rabbits. (Oboh, et al., 2007). The consumption of high protein can negatively impact the overall cholesterol level by causing a significant reduction in it. (Oboh, & Olumese, 2008).

According to the findings, Group C (29.00±1.32 mg/dL) demonstrates elevated levels of HDL cholesterol in comparison to Group A (4.80±0.14 mg/dL) and Group B (16.00±1.65 mg/dL). Earlier research conducted by another scholar has also suggested that the consumption of high protein is linked to an increase in HDL cholesterol levels. (Oboh, & Olumese, (2008).

Based on the findings, it appears that Group C (34.00±3.12 mg/dL) has higher LDL cholesterol levels compared to Group A (32.00±0.97 mg/dL) and Group B (4.50±0.84 mg/dL). Similar experiments have been conducted, indicating that consuming a carbohydrate diet can decrease LDL cholesterol levels. (Oboh, et al., 2007).

According to the findings, Group B (27.83±3.10 mg/dL) displayed higher levels of triacylglycerol compared to Group A (10.28±0.66 mg/dL) and Group C (18.00±0.96 mg/dL). Similar research has indicated that triacylglycerol levels tend to increase when subjects are fed a carbohydrate-rich diet. (Oboh, et al., 2007). Human subjects who were provided with a highly regulated carbohydrate-rich diet were also analyzed in similar studies. (Katibi et al., 2004)

The results show that group B (0.8400±0.0603 mg/dL) has high values of bilirubin levels compared to group A (0.2400±0.0390 mg/dL) and group C (0.2800±0.0493 mg/dL). similar studies indicate that a carbohydrate-rich diet increases blood bilirubin levels (Oloruntola, et al., 2018). some studies indicate that the carbohydrate diet does not affect the blood bilirubin level (Oboh, et al., 2007).

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