Uttar Pradesh Journal of Zoology

Volume 45, Issue 6, Page 79-88, 2024; Article no.UPJOZ.3331
ISSN: 0256-971X (P)

Biochemical Parameters of Female Wistar Rats Fed Shellfish and Cabbage as Potential Nutritive Supplements for Malnutrition

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

Article Information

DOI: 10.56557/UPJOZ/2024/v45i63952

Open Peer Review History:
This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://prh.mbimph.com/review-history/3331

Original Research Article

Received: 07/01/2024
Accepted: 11/03/2024
Published: 15/03/2024

ABSTRACT

Inability to meet the nutritional requirements causes malnutrition which further leads to various disorders and makes people susceptible to infections. Malnutrition is a serious problem globally and this research was conducted with an aim to understand the nutritive potentials of Shellfish and Cabbage to help deal with malnutrition. Female Wistar rats in post weaning stage were placed in six different groups on the basis of diets provided for a duration of 20 weeks. Investigations for biochemical parameters such as Total Protein, Albumin, enzymes Aspartate aminotransferase (AST), Alanine aminotransferase (ALT), Alkaline phosphatase (ALP), Bilirubin, Creatinine, Blood Urea Nitrogen, Cholesterol and Triglycerides were done to determine the potentials of these food sources as dietary supplements.

Keywords: Nutritive supplements; cabbage; shellfish; total protein; triglycerides.

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1. INTRODUCTION

Malnutrition is the condition in which a person has deficiency or excess of nutrients. Accordingly, malnutrition includes undernutrition involving deficiency of nutrients and also conditions such as being overweight and obese. Malnutrition can be both, a cause of the disease as well as a consequence of a disease [1]. Women are more likely to suffer from nutritional deficiencies than men are, for reasons including women's reproductive biology, low social status, poverty and lack of education [2]. India is known to have one of the highest number of malnourished children and upto a million children die before one month of age as many young mothers are anemic. Mortality rates of both mothers as well as children due to malnutrition are considerably high [3]. During early age, as the development of brain as well as body as a whole is at its peak, undernutrition in childhood is definitely going to leave a long lasting mark on an individual's development and body functioning [4]. It is known that proper nutrition is essential in achieving a satisfactory level of human development [5].

Malnutrition affects biochemical parameters negatively. Hypoinsulinemia and discrete hypoglycemia were found in adult Wistar rats, due to protein restriction during lactation [6,7]. Significant decreases in total protein concentration in mice have also been observed [7-10]. Changes in blood sugar, insulinemia and leptinemia in Wistar rats, caused by protein restriction during gestation and/or lactation, can be passed transgenerationally to the second generation of pups [7, 11]. Protein malnutrition reduces the content of many proteins in the body, including enzymes and hormonal proteins [12,13]. Physicians more often rely on albumin levels to assess a patient's nutritional status [13,14]. Albumin concentration is a better predictor of underlying malnutrition in the cardiac transplant recipients than the body mass index [13,15]. It is also a better prediction factor in the evaluation of malnutrition in patients undergoing elective orthopaedic surgery [13,16]. A lower concentration of albumin may be also related to the liver function damage [13,17]. Malnutrition is known to affect liver cells and causes liver enzyme imbalances. Several studies have shown that malnutrition can increase the levels of liver enzymes such as ALT and AST in patients. Such changes have been reported in different degrees of malnutrition. The amount of ALP was found to be lower in malnourished children [18-20].

Shellfishes are an important source of high quality animal protein [21]. Shellfishes also have lipids, glycogen and minerals that contribute to the nutritional value of their soft tissues. These molecules along with minerals and minor components of lipophilic and hydrophilic nature, contribute to its organoleptic characteristics and nutritional value [22]. Protein is the most abundant biochemical component in tissues and in some Shellfishes, it may be an alternative energy reserve during gametogenesis [23]. Many studies have suggested that increasing consumption of plant foods like Cabbage decreases the risk of obesity, diabetes, heart disease and overall mortality while promoting a healthy complexion, increased energy and overall lower weight. Cabbage contains beta-carotene, lutein and zeaxanthin, carotenoids that are a large class of natural plant pigments. They have chemo-protective effects, exhibit strong antioxidant properties and may reduce the risk of age related macular degeneration and some types of cancer [24-26]. Finding solutions to deal with the problem of malnutrition is the need of the hour. This paper focuses on the biochemical parameters of Wistar rats fed Shellfish and cabbage as potential nutritive supplements to help overcome malnutrition.

2. MATERIALS AND METHODS

After obtaining the required approval from Institutional Animal Ethics Committee (IAEC), 36 female Wistar rats in post weaning stage, obtained from Bharat Serums and Vaccines, Thane were brought to the Animal house at Dr. L.H. Hiranandani College of Pharmacy, CHM College Campus, Ulhasnagar. The rats were housed in Polypropylene cages with corn cob bedding. Rats were acclimatized for two weeks. They were provided normal rat feed pellets obtained from Nutrivet Lifesciences, Pune *ad libitum* during the acclimatization period. Water was provided *ad libitum* to all the animals throughout the experiment duration. After two weeks, rats were divided into six groups randomly, with six rats in each group. They were fed group specific diet as: Group 1 rats were fed Normal rat feed pellets *ad libitum* throughout the experiments. Rats in Group 2 were fed Normal rat feed pellets for 75% of their dietary requirements and remaining 25% was Shellfish. Group 3 rats were fed Normal rat feed pellets for 75% of their dietary requirements and remaining 25% was Cabbage. Group 4 rats were malnourished as they were on a restricted diet, being fed just 50% of their dietary requirement of rat feed pellets. Group 5 rats were also malnourished being fed just 50% of their dietary requirement of rat feed pellets but these were provided with Shellfish as dietary supplement for the remaining 50% of their dietary requirements. Rats in Group 6 were malnourished too, being fed just 50% of their dietary requirement of rat feed pellets and these were provided with Cabbage as dietary supplement for the remaining 50% of their dietary requirements.

Feed consumption of Group 1 rats (*ad libitum*) was noted daily and it served the basis for calculation of feed requirements for the other groups. Shell fish (Squid) and Green Cabbage were obtained from local market, cleaned with water, cut into small pieces, weighed and fed to the rats as per their feeding groups. The feeding experiments were carried out for 20 weeks after which the rats were sacrificed using CO_2 chamber. Blood samples to study biochemical parameters were collected by cardiac puncture method and serum was used for the biochemical tests. The parameters investigated were Bilirubin (Modified Jendrassik and Grof's method), Aspartate aminotransferase (Modified Reitman and Frankel's Colorimetric DNPH method), Alanine aminotransferase (Reitman and Frankel's Colorimetric DNPH method), Alkaline Phosphatase (p-NPP kinetic method), Total protein (Biuret method), Albumin (BCG dye binding method), Globulin (Calculated using Total protein and Albumin values), Blood Urea Nitrogen (calculated using Urea estimated by Berthelot method), Creatinine (Modified Jaffe's method), Random Blood Sugar (GOD POD method), Cholesterol (Trinder CHOD/PAP method), Triglycerides (GPO/PAP method).

2.1 Statistical Analysis

Minitab Software was used to analyze the result by one way analysis of variance (ANOVA) to determine the differences between the mean values at $P < 0.05$ level followed by Tukey post hoc test. Data presented as Mean ± SD. Graphs were created using Microsoft Excel.

3. RESULTS AND DISCUSSION

The Bilirubin (Total) content did not show any significant difference among any of the groups. With respect to AST (Aspartate aminotransferase) enzyme, Group 5 and 6 showed significant difference compared to group 1(Normal rat feed). Highest AST levels were shown by group 1 and lowest by group 6. Normal rat feed group 1 had highest while Groups 4 and 6 showed lowest levels of ALT (Alanine aminotransferase) enzyme, however, it was statistically insignificant. Group 6 rats which were malnourished and fed Cabbage showed lowest level of ALP (Alkaline Phosphatase) enzyme and it was significantly different as compared to group 3 rats with normal feed and Cabbage which showed highest value of ALP. Total Protein content in malnourished rats (group 4) was significantly lower to groups 1 and 2 while that of group 5 was significantly higher from group 4. Both the Shellfish fed groups showed higher level of total protein. Albumin content of groups 4 and 6 was significantly lower compared to groups 1, 2 and 3 while that of group 5 malnourished rats being fed Shellfish was

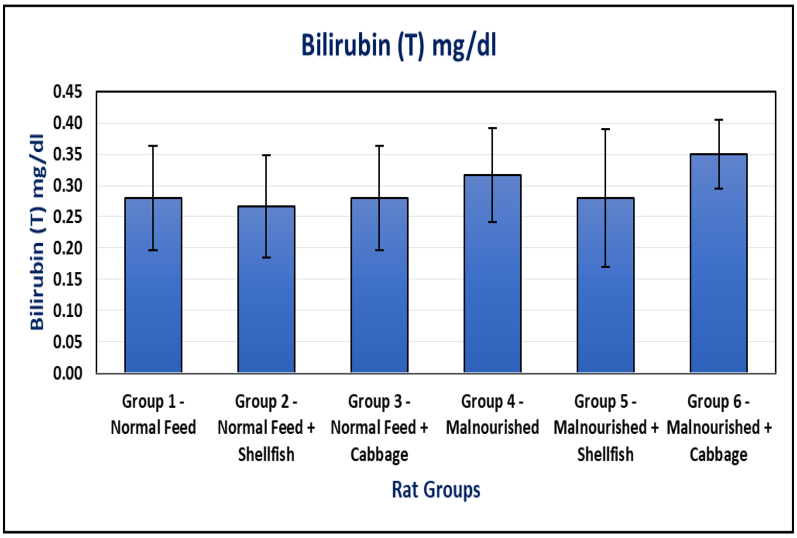

Fig. 1. Graph of Bilirubin (T) (Mean ±SD) vs Rat groups

Table 1. Biochemical parameters

Rat Groups	Bili (T) mg/dl	AST (IU/L)	ALT (IU/L)	ALP (IU/L)	Total Protein (g/dl)	Albumin (g/dl)	Globulin (g/dl)	Creatinine (mg/dl)	RBS (mg/dl)	BUN (mg/dl)	Cholesterol (mg/dl)	Triglycerides (mg/ dl)
Group 1 - Normal Feed	0.28 ± 0.08	222.4 ± 37	148.2 ± 28.00	235.80 ± 103.00	7.04 ± 0.21	3.40 ± 0.12	3.64 ± 0.15	0.62 ± 0.08	84.40 ± 14.88	17.24 ± 1.58	51.40 ± 8.26	177.60 ± 20.28
Group 2 - Normal Feed + Shellfish	0.27 ± 0.08	183.80 ± 46.20	135.80 ± 37.60	268.30 ± 82.50	7.15 ± 0.21	3.42 ± 0.16	3.60 ± 0.09	0.65 ± 0.05	95.00 ± 10.55	16.72 ± 1.84	54.50 ± 4.85	191.67 ± 16.17
Group 3 - Normal Feed + Cabbage	0.28 ± 0.08	178.60 ± 22.60	120.40 ± 21.52	377.20 ± 118.30	6.96 ± 0.23	3.42 ± 0.13	3.54 ± 0.18	0.58 ± 0.08	93.80 ± 20.97	16.32 ± 0.73	48.00 ± 4.06	193.80 ± 15.06
Group 4 - Malnourished	0.32 ± 0.08	171.33 ± 23.47	118.83 ± 14.12	286.70 ± 70.50	6.42 ± 0.48*	3.00 ± 0.14*	3.42 ± 0.38	0.57 ± 0.05	80.17 ± 12.86	17.72 ± 1.28	50.17 ± 11.09	153.00 ± 35.50
Group 5 - Malnourished + Shellfish	0.28 ± 0.11	162.00 ± 21.76*	123.20 ± 20.61	250 ± 112.30	7.08 ± 0.41*	3.36 ± 0.21*	3.72 ± 0.41	0.60 ± 0.07	87.20 ± 7.63	17.2 ± 1.26	51.80 ± 10.26	181.60 ± 37.10
Group 6 - Malnourished + Cabbage	0.35 ± 0.05	152.80 ± 28.80*	118.80 ± 29.10	173.70 ± 40.00*	6.72 ± 0.43	2.97 ± 0.27*	3.75 ± 0.34	0.48 ± 0.04*	86.67 ± 9.35	19.77 ± 1.12*	49.00 ± 7.56	135.00 ± 19.41*

*Data presented as Mean ± SD; *Means are statistically significant at P<0.05 (One way ANOVA followed by Tukey post hoc test). Bili (T)- Total Bilirubin, AST – Aspartate Aminotransferase, ALT- Alanine aminotransferase, ALP- Alkaline Phosphatase, RBS- Random Blood Sugar, BUN – Blood Urea Nitrogen*

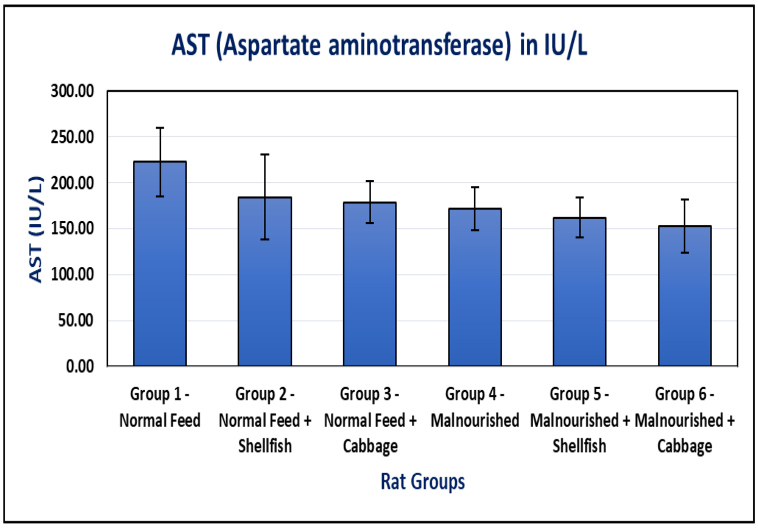

Fig. 2. Graph of AST (Mean ±SD) vs Rat groups

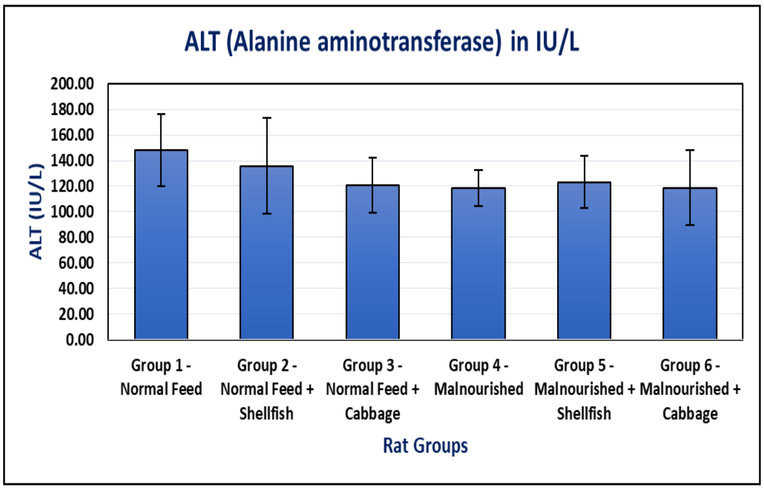

Fig. 3. Graph of ALT (Mean ±SD) vs Rat groups

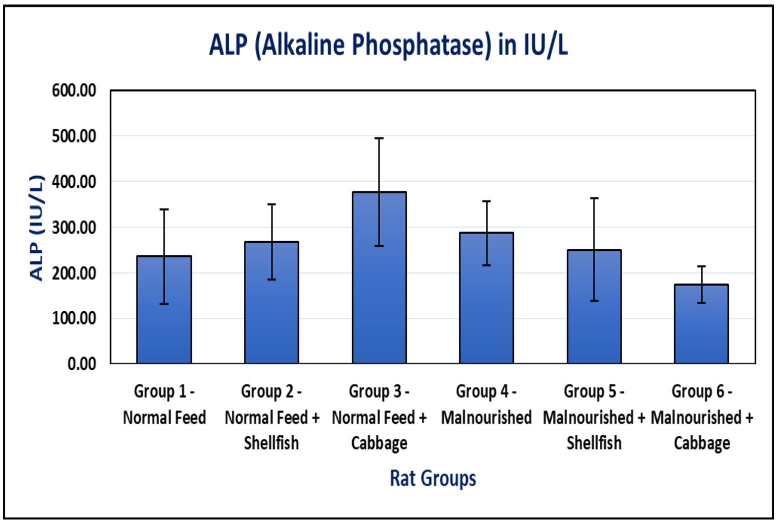

Fig. 4. Graph of ALP (Mean ±SD) vs Rat groups

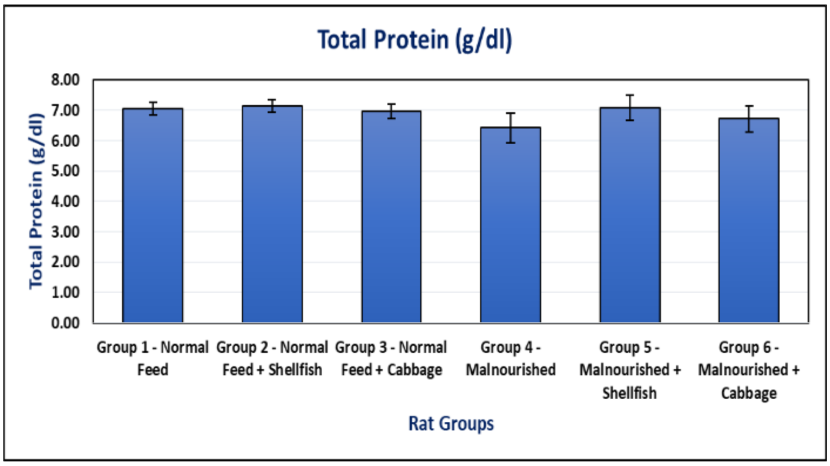

Fig. 5. Graph of Total protein (Mean ±SD) vs Rat groups

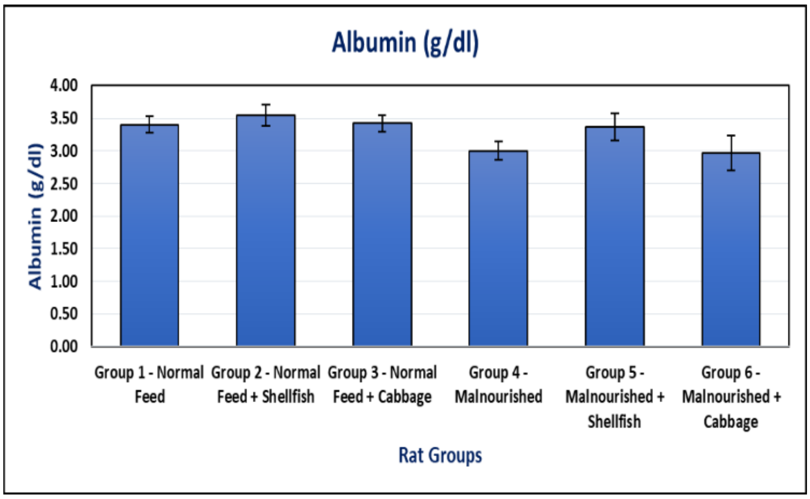

Fig. 6. Graph of Albumin (Mean ±SD) vs Rat groups

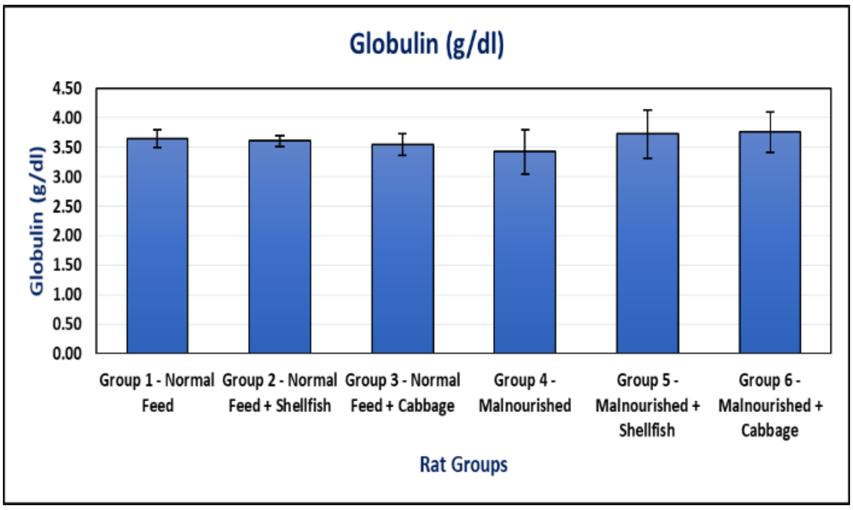

Fig. 7. Graph of Globulin (Mean ±SD) vs Rat groups

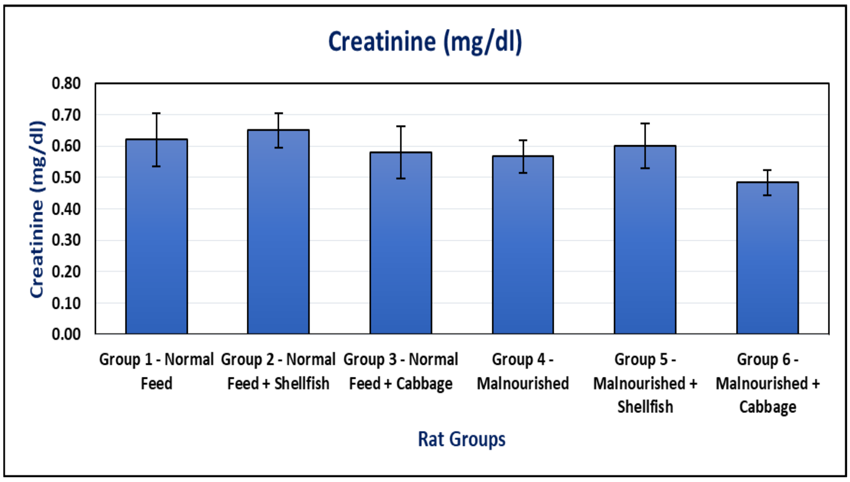

Fig. 8. Graph of Creatinine (Mean ±SD) vs Rat Groups

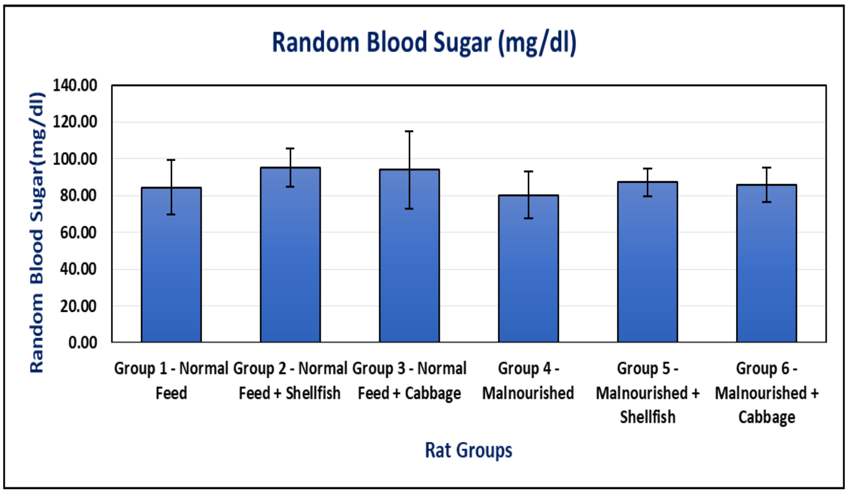

Fig. 9. Graph of random blood sugar (Mean ±SD)

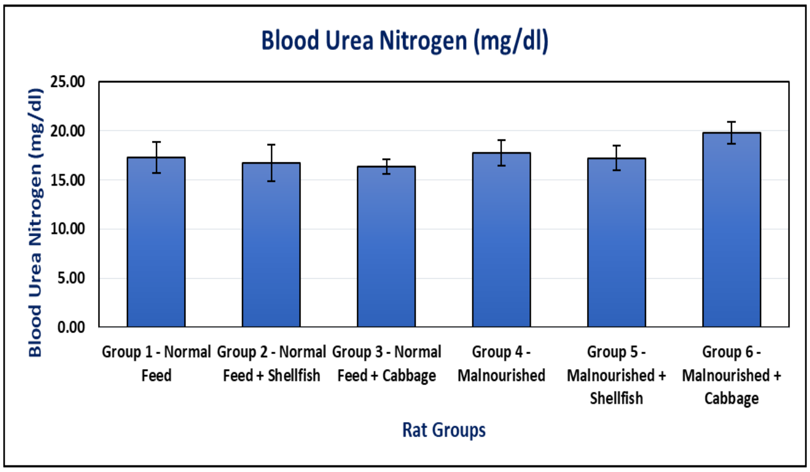

Fig. 10. Graph of blood urea nitrogen (Mean vs Rat groups ± SD) vs Rat Groups

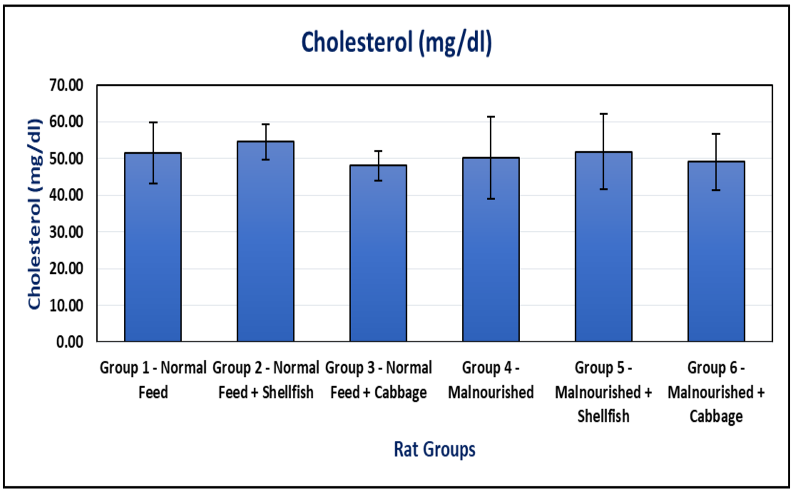

Fig. 11. Graph of Cholesterol (Mean ±SD) vs Rat groups

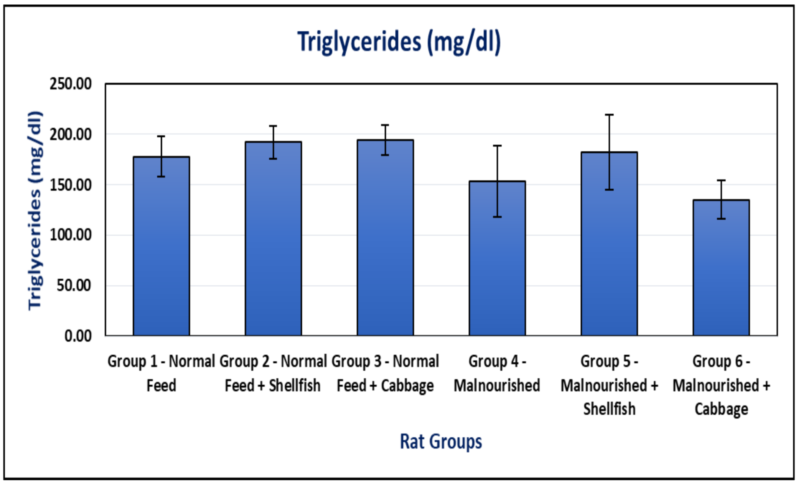

Fig. 12. Graph of Triglycerides (Mean ±SD) vs Rat groups

significantly higher compared to group 4. No statistically significant result observed with respect to Globulin. Creatinine levels of Group 6 were lowest and significantly different from groups 1 and 2. Random Blood sugar was highest for group 2 and lowest for group 4, no statistically significant difference was observed. Blood Urea Nitrogen level was highest in group 6 and statistically significant in comparison to groups 1, 2 and 3. It was lowest in group 3. Both Shellfish fed groups i.e. group 2 and 5 showed higher Cholesterol levels compared to normal feed group but no statistically significant difference was observed. Triglyceride levels were lowest in group 6 and showed significant difference compared to groups 1, 2 and 3. Highest level of Triglycerides was observed in group 3.

4. CONCLUSION

In this study, Shellfish and Cabbage reduced the levels of enzymes Aspartate aminotransferase and Alkaline Phosphatase when given as nutritive supplements to malnourished rats. Shellfish diet for malnourished rats significantly increased the Total protein as well as Albumin content. Cabbage diet given to malnourished rats was found to significantly increase Blood Urea nitrogen and significantly lowered Creatinine and Triglyceride levels.

ETHICAL APPROVAL

Animal Ethic committee approval has been collected and preserved by the author(s).

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history:
The peer review history for this paper can be accessed here:
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