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EXPLORING THE THERAPEUTIC POTENTIAL OF *RAPHANUS SATIVUS* LEAF PROTEIN CONCENTRATE: AN IN-DEPTH STUDY USING WISTAR RAT AS ANIMAL MODEL

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ABSTRACT

Raphanus sativus is a well-known root vegetable crop belonging to the Brassicaceae family. *Raphanus sativus* (radish) is a source of nutrients and phytochemicals, particularly proteins, glucosinolates, flavonoids, β-carotene, and minerals. This investigation explores the dietary effects of leaf protein concentrate (LPC) made from *Raphanus sativus* leaves, focusing on the protein energy ratio (PER), feed efficiency ratio (FER) and the impact on biochemical parameters in Wistar rats. *Raphanus* leaves, being a potential source of plant-based protein, were processed into LPC and incorporated into the diet of wistar rats. The study aimed to assess the acute toxicity, protein quality and overall nutritional value of LPC through the calculation of PER and FER. Furthermore, the investigation extended to evaluating the influence of LPC consumption on various biochemical parameters in the blood serum of the rats.

KEYWORDS: Leaf Protein Concentrate (LPC), *Raphanus sativus*, Protein Energy Ratio (PER), Feed Efficiency Ratio (FER), biochemical parameters, Acute toxicity.

INTRODUCTION

Raphanus sativus is a well-known root vegetable crop belonging to the Brassicaceae family. (Manivannan et al., 2019). *Raphanus sativus* (radish) is a source of nutrients and phytochemicals, particularly proteins, glucosinolates, flavonoids, β-carotene, and minerals. (Gamba et al., 2021).

To comprehensively evaluate the protein quality and safety of raw or processed food materials intended for human consumption, it is common practice to conduct testing on animal models, which can yield pertinent information. (Adeoti oluwole A et al, 2018). There is a need to assess the safety of plant extracts for human consumption prior to considering their potential therapeutic role. One of the effective ways by which this can be done is through conducting acute oral toxicity tests *in vivo*. Acute toxicity is characterized by unfavorable effects that occur either immediately or within a specific time frame following the administration of single or multiple doses of a substance .(Muñoz et al., 2021).

MATERIALS AND METHOD

The fresh leaves of *Raphanus sativus* were collected from Bhandup vegetable market, Bhandup east, Mumbai, Maharashtra. Leaf protein concentrate (LPC) was made as per the method described by Fellows, 1987; and was used to carry out further analysis. For acute toxicity, OECD 425 (2022) was followed and the protein deficiency tests were as per Sadasivam, 2008. This study was approved by the Institutional Animal Ethics Committee (Reg. no.- CPCSEA/IAEC/P-73/2022). The male rats were divided randomly into two groups consisting of five rats. Animals of group I (Control) received normal diet and group II – experimental group, received LPC of *Raphanus sativus* and normal diet for twenty-eight consecutive days. Recorded the food consumption and body weights at weekly intervals.

The experimental design involved feeding the Wistar rats with diets containing LPC and monitoring their growth (in terms of body weight), feed intake, and protein utilization. PER, a fundamental measure of protein quality, was determined by comparing weight gain to protein intake. FER, indicative of the efficiency of nutrient utilization, was also calculated by relating weight gain to feed intake. Simultaneously, blood serum samples were analysed for biochemical parameters. These included the examination of LPC of *Raphanus sativus*, as a potential source of nutritional support.

Statistical analysis - All data are expressed as mean and standard deviation (SD). The evaluation of statistical significance was performed using Student t-test. The criterion for statistical significance is expressed as $p < 0.05$.

RESULTS

Acute oral toxicity study- The treated animals that received the largest oral dose of LPC of *Raphanus sativus* i.e., 2000 mg/kg, lived through the entire trial and showed no symptoms up to 14 days following dosing.

Table 1: PER and FER results of *Raphanus sativus* (LPC).

Parameters	Control	Experimental (LPC)
Initial weight (g)	100.30 ± 15.28	90.62 ± 5.63
Final weight(g)	224.3 ± 23.01	203.13 ± 12.55
Weight gain (g)	123	124
FER	0.84	0.84
PER	2.19	2.21

mean ±SD (n=5). Average weight gain ± SD.

No significant variation observed in between control and LPC of *Raphanus sativus* which is ($p < 0.05$).

Biochemical parameters- For the serum biochemistry profile analysis, blood samples without anticoagulants were used. Table 2 shows the mean Total protein, Albumin, ALT (Alanine Aminotransferase), AST (Aspartate Aminotransferase), ALP (Alkaline Phosphatase), and Creatinine levels.

Table 2: Biochemical parameters of LPC of *Raphanus sativus*.

	Total Protein g /dl	Albumin g / dl	ALT IU/L	AST IU/L	ALP IU/L	Creatinine mg/dl
Control	6.1 0± 1.08	2.9± 0 17	81±3.60	62.6±2.51	45.3±3.05	0.6±0.05
Experimental (LPC)	6.26 ±0.05	2.83± 0.05	95±3.60	64± 3	44.6±2.51	0.7±0.1

Mean ± SD (n=5).

Table 2 presents data for the control group and the experimental group across various biochemical parameters. The experimental group showed no significant difference compared to the control group in terms of total protein, albumin, AST, and ALP. However, there is a difference in ALT levels, with the experimental group having higher ALT levels. Additionally, the creatinine levels are higher in the experimental group but within the normal range.

There was no significant difference in ALT, AST, ALP, and Creatinine serum levels between the treated and control groups. Our findings indicate that the LPC of *Raphanus sativus* did not cause any liver or kidney damage.

DISCUSSION

Halimatul et al., 2007 studied the protein quality of *Hibiscus Sabdariffa* L. seeds. Mhlomi et al., 2022 conducted the assessment of rats fed protein-deficient diets supplemented with *Moringa oleifera* leaf meal. Clinical diagnosis of disease and damage to the structural integrity of the liver is commonly assessed by monitoring the status of serum AST, ALP and ALT activities, which are sensitive serological indicators of liver toxicity. (Cui et al., 2011). When there is an injury to the organs due to any reason then these enzymes are spilled into the bloodstream. Creatinine is a chemical waste product that is carried via the bloodstream and eliminated by the kidneys. If the filtration in the kidney is deficient, creatinine blood levels rise (Hasan et al., 2017). The reference ranges of AST, ALT and ALP, total protein, creatinine, and albumin are 50 to 150 IU/L, 52 to 224 IU/L, 30 to 130 IU/L,4.5- 8.4 mg/dl,0.4-1.4 mg/dl and 2.9-5.9 g/dl respectively. (Sharp and Villano, 2013).

CONCLUSION

In conclusion, this study has provided valuable insights into the nutritional and physiological implications of incorporating *Raphanus sativus* (LPC) into diets. The assessment of the Protein Energy Ratio (PER) and Feed Efficiency Ratio (FER) has illuminated the protein quality and utilization efficiency of LPC, reinforcing its potential as a sustainable and viable protein source. The calculated PER values, which reflect the ratio of weight gain to protein intake, demonstrate that *Raphanus* leaf-derived LPC holds promise as a valuable protein supplement. Furthermore, the evaluation of blood serum biochemical parameters has offered a comprehensive understanding of the potential physiological effects of LPC consumption. These findings underscore the need for continued investigation into the broader health implications of incorporating LPC into human diets.

The utilization of *Raphanus* leaves for LPC production not only contributes to addressing protein deficiencies but also aligns with sustainability goals by repurposing plant parts that might otherwise go to waste. By capitalizing on this readily available and underutilized resource, we can work towards more efficient and ecologically sound nutritional supplements production.

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