

## ORIGINAL ARTICLE

## COMPARISON OF SERUM LIPID PARAMETERS IN NORMAL, DIABETIC AND TURMERIC TREATED DIABETIC RATS

Javaria Latif, Aysha Zaheer\*, Mah e Jabeen Sear\*\*, Saima Mukhtar\*\*, Iram Qamar\*\*, Sara Naeem\*\*

Department of Physiology, CMH Institute of Medical Sciences, Bahawalpur, \*Services Institute of Medical Sciences, Lahore, \*\*Rahbar Medical &amp; Dental College, Lahore, Pakistan

**Background:** Alteration in lipid profile is a common complication of diabetes leading to life threatening conditions. Aim of this study was to compare serum lipid profile parameters in control, diabetic and diabetic animals given turmeric powder and to determine the improvements in lipid profile in diabetic rats with the use of turmeric powder. **Methods:** In this experimental study, carried out at Postgraduate Medical Institute Lahore, 45 albino rats were randomly divided into three equal groups (normal control, diabetic and diabetic receiving turmeric therapy). Initially, Alloxan was administered to all the animals except normal control to produce diabetes. After confirmation of diabetes, one diabetic group was selected to be treated with turmeric powder dissolved in water in dosage of 250 mg/Kg body weight. Blood samples were obtained from rats after 12-hour fasting period at the start of experiment after confirmation of diabetes, week eight and week twelve. **Results:** When comparison was made, mean values of lipid profile parameters were significantly higher for diabetic control and diabetic animals receiving turmeric therapy as compared to normal control during week eight but the difference between both the diabetic groups was also significant. By the end of week twelve the difference between normal control and diabetic animals receiving turmeric therapy became non-significant with improvement in lipid profile parameters of diabetic rats receiving turmeric powder. **Conclusion:** Therapy with turmeric powder induces improvement in lipid profile of diabetic rats with dyslipidemia.

**Keywords:** Curcumin, Diabetes Mellitus, Lipid parameters

Pak J Physiol 2025;21(4):26–9, DOI: <https://doi.org/10.69656/pjp.v21i4.1874>

## INTRODUCTION

According to the report of ‘International Diabetes Federation’ approximately 537 million individuals in between 20 to 79-year of age are having diabetes, expected to rise by 783 million in 2045.<sup>1</sup> Middle East and South East Asia are becoming high disease prevalent countries due to sedentary life styles, obesity and unhealthy diets.<sup>2,3</sup>

Alteration in lipid profile parameters (dyslipidemia) is common in diabetics, exhibiting poor glycemic control.<sup>4,5</sup> Deficiency of insulin and impaired insulin sensitivity are the reasons to develop altered lipid metabolism.<sup>6</sup> Increased levels of total serum cholesterol, serum phospholipids and low serum HDL cholesterol are major risk factors for development of cardiovascular complications in diabetic individuals.<sup>7</sup> By providing proper diet regimen, making appropriate lifestyle changes including exercise and keeping the stability of blood glucose levels in normal range, complications associated with diabetes (dyslipidemia) can be prevented.<sup>7,8</sup>

Turmeric plant since primordial times is found to have medicinal properties and is commonly available in tropical (Asian) countries.<sup>9</sup> According to the recent research Curcumin, an active ingredient of turmeric is helpful to maintain normal blood lipid levels by controlling the absorption, transport and excretion of cholesterol.<sup>10</sup> Turmeric is believed to maintain

cholesterol homeostasis by increasing the expression of enzyme CYP7A1, which is involved in cholesterol metabolism.

Through its anti-inflammatory and anti-oxidant effects, turmeric (curcumin) is able to prevent development of intractable complications induced by diabetes like atherosclerosis and ischemic heart disease.<sup>11</sup> By reducing the levels of inflammatory indicators (hs-CRP, TNF- $\alpha$  and IL-6), curcuminoids have protective effect on the beta cells of pancreas and improvement in insulin resistance thus beneficial to maintain blood glucose levels of diabetics and indirectly preventing the abnormal alterations in serum lipid levels.<sup>12</sup>

This study was focused to compare some of main serum lipid profile parameters among normal, untreated diabetic, and diabetic rats on turmeric therapy, and to find out some dietary supplement which can reduce the chances of having complications like dyslipidemia in diabetic patients.

## MATERIAL AND METHODS

Forty-five Sprague-Dawley rats with average age of 11 weeks, were selected for this study. Sample size was calculated using resource equation method for one-way ANOVA by using the formula  $n = DF/k + 1$  to obtain maximum number of animals per group. Rats were acclimatized in animal house of Post-Graduate Medical Institute, Lahore for one week. All the rats had free

access to the rat chow diet and water. Animals were kept at  $24\pm 5$  °C with 12-hour light/dark cycle. Total duration of this experiment was 4 months, approval was granted by Ethical Committee of Animal Sciences of the Institute.

After completion of acclimatization period, all 45 rats were randomly divided into 3 equal groups of 15 using simple random sampling with balloting method. All animals except normal controls were administered intraperitoneal injection of alloxan 130 mg/Kg body weight as one-time dose, dissolved in normal saline after overnight fast. Diabetes was confirmed in animals after 5–7 days of alloxan injection on tail vein blood with glucometer. Animals having blood glucose levels  $>200$  mg/dL were included in the experiment.

For the next 12 weeks, healthy rats of normal control group and the animals of diabetic control group were given normal synthetic rat diet and 3 mL distilled water per day. The third group animals were given powdered rhizomes of *Curcuma longa* (turmeric) 250 mg/Kg body weight per day dissolved in 3 mL of distilled warm water.

Blood samples (1.5–2.5 mL per animal) of all animals were taken from saphenous vein after a fasting period of 12 hours. Blood was collected initially when diabetes was confirmed, then after 8 weeks and finally by the end of 12<sup>th</sup> week.<sup>13</sup> Total serum cholesterol, total serum phospholipids, LDL-c and HDL-c were measured by direct quantitative (enzymatic colorimetric method).<sup>14</sup> Automated chemistry analyser was used for tests performed in Chemical Pathology Lab of PGMI Lahore.

Data analysis was done using SPSS-23. Quantitative data were expressed as Mean $\pm$ SD. To compare lipid profile parameters among groups, one-way ANOVA was used. For comparison of all the possible pairs of means, Post hoc Tukey test was applied, and  $p\leq 0.05$  was considered statistically significant.

## RESULTS

At the start of experiment, mean total serum cholesterol level for animals in healthy control group was  $73\pm 7$  mg/dL, which remained in normal range during twelve-week time period and was  $64\pm 7$  mg/dL at the end. The total serum cholesterol level for diabetic group and diabetic animals having turmeric therapy, increased to  $143\pm 13$  mg/dL and  $131\pm 15$  mg/dL respectively, after induction of diabetes. The total serum cholesterol level of rats receiving turmeric declined during experiment with mean total serum cholesterol level  $91\pm 12$  mg/dL at week 8 and  $64\pm 19$  mg/dL at last day of study. An increase in total serum cholesterol level for diabetic control animals was recorded with mean total serum cholesterol level  $214\pm 22$  mg/dL at the end (Table-1).

When comparison was made among groups at different times, it was observed that mean total serum cholesterol levels were significantly different ( $p<0.001$ ).

By the end of week eight, diabetic control animals and those having turmeric therapy had significantly higher mean values ( $p<0.001$ ) as compared to normal control, and the difference between the diabetic control and turmeric receiving diabetics was also significant ( $p<0.001$ ). Mean total serum cholesterol level of diabetic control animals raised significantly as compared to normal control ( $p<0.001$ ) at the end of the experiment and was also significantly higher from the diabetic animals receiving turmeric powder ( $p<0.001$ ). The difference between normal control and diabetics having turmeric treatment was found insignificant ( $p=0.987$ ), at the end of week 12. (Table-1).

The mean total serum phospholipids level of animals in normal control group was  $103\pm 8$  mg/dL at the start and  $92\pm 7$  mg/dL at the end. The serum phospholipids level increased to  $110\pm 11$  mg/dL and  $126\pm 14$  mg/dL for the other two groups after diabetes. The serum phospholipids level gradually decreased for animals receiving turmeric throughout the experiment with mean value  $90\pm 15$  mg/dL by the end of week 12. Gradual increase in serum phospholipids level of diabetic control animals was observed with mean serum phospholipids level  $129\pm 21$  mg/dL and  $170\pm 38$  mg/dL at week 8 and 12. (Table-2).

The pair-wise comparison explained a higher mean value in turmeric treated animals as compared to the other two groups ( $p<0.001$  respectively) and the difference between normal control and diabetic control was not significant ( $p=0.297$ ) at the start of study. At week 8 the diabetic control had significantly higher average of total serum phospholipids by  $31.4$  mg/dL and  $26.9$  mg/dL ( $p<0.001$ ) as compared to normal control and animals receiving turmeric therapy. By the end of week 12, mean serum phospholipids level of animals in diabetic control increased by  $78.5$  mg/dL as compared to normal control ( $p<0.001$ ) and was significantly higher by  $80.9$  mg/dL from diabetic treated animals ( $p<0.001$ ). The difference of  $2.4$  mg/dL between diabetics and diabetics with turmeric treatment was recorded which was insignificant ( $p=0.957$ ).

At the start of experiment, the mean value of LDL/HDL ratio observed for animals in healthy control group was  $2.83\pm 0.60$ , which declined a bit during 12-week time and was  $2.44\pm 0.49$  at the end. The LDL/HDL ratio for animals in other two groups increased to  $5.80\pm 1.78$  and  $7.72\pm 4.69$  respectively after diabetes. The LDL/HDL ratio of animals declined for diabetic animals having turmeric treatment during the experiment with mean value of  $4.53\pm 2$  at week 8 and  $2.62\pm 1$  at the end. The LDL/HDL ratio for diabetic control animals was observed to be increased with mean LDL/HDL ratio  $11.06\pm 5.7$  at week 8 and  $19.18\pm 6.9$  at week 12. When groups were compared at different measurement times, the mean ratios were found statistically significant ( $p<0.001$ ). (Table-3).

Initial pair-wise comparison done by post-hoc Tukey test showed a higher mean value of LDL/HDL ratio in diabetic control animals and those receiving turmeric when compared to healthy control animals ( $p < 0.05$  and  $< 0.001$  respectively). The diabetic control animals had significantly higher average ratio ( $p < 0.001$ ) as compared to the other two groups by week 8, and the difference between the normal control group and diabetic turmeric treated group was not significant ( $p = 0.319$ ). By the end of week 12, the mean LDL/HDL ratio of diabetic control animals was higher by 16.74 compared to normal control ( $p < 0.001$ ) and was significantly higher by 16.56 from rats having turmeric therapy ( $p < 0.001$ ). The difference of 0.18 between normal control and turmeric receiving diabetic animals was observed which was not statistically significant ( $p = 0.991$ ).

**Table-1: Comparison of total serum cholesterol levels of animals at given times (mg/dL, Mean±SD)**

| Animal Groups             | start of study (after diabetes) | 8 <sup>th</sup> week | 12 <sup>th</sup> week |
|---------------------------|---------------------------------|----------------------|-----------------------|
| Healthy control           | 73±7                            | 69±6                 | 64±7                  |
| Diabetic control          | 143±13                          | 185±15               | 214±22                |
| Diabetic+turmeric treated | 131±15                          | 91±12                | 64±19                 |
| <i>p</i>                  | <0.001                          | <0.001               | <0.001                |

**Table-2: Comparison of serum phospholipids levels of animals at given times (mg/dL, Mean±SD)**

| Animal groups             | start of study (after diabetes) | 8 <sup>th</sup> week | 12 <sup>th</sup> week |
|---------------------------|---------------------------------|----------------------|-----------------------|
| Healthy control           | 10±8                            | 98±9                 | 92±7                  |
| Diabetic control          | 110±11                          | 129±21               | 170±38                |
| Diabetic+turmeric treated | 126±14                          | 102±17               | 90±15                 |
| <i>p</i>                  | <0.001                          | <0.001               | <0.001                |

**Table-3: Comparison of LDL/HDL-cholesterol ratio of animals at given times (mg/dL, Mean±SD)**

| Animal groups             | start of study (after diabetes) | 8 <sup>th</sup> week | 12 <sup>th</sup> week |
|---------------------------|---------------------------------|----------------------|-----------------------|
| Healthy control           | 2.83±0.60                       | 2.66±0.70            | 2.44±0.49             |
| Diabetic control          | 5.80±1.78                       | 11.06±5.70           | 19.18±6.99            |
| Diabetic+turmeric treated | 7.72±4.69                       | 4.53±2.01            | 2.62±1.03             |
| <i>p</i>                  | <0.001                          | <0.001               | <0.001                |

## DISCUSSION

This study's findings emphasize the notable dyslipidemia linked to diabetes and illustrate the possibility of turmeric powder as a therapeutic measure to alleviate these metabolic disorders. The enhanced blood lipid profile characteristics reported in diabetic control rats at week eight correspond with established data that hyperglycaemia intensifies lipid irregularities by facilitating lipolysis, augmenting free fatty acid flow, and disrupting lipoprotein metabolism.<sup>15</sup> Notably, the diabetic group receiving turmeric exhibited a significant reduction in lipid parameters compared to the untreated diabetic group by week eight, suggesting an early but partial therapeutic effect. However, the normalization of lipid profiles in the turmeric-treated group by week 12, were close to the non-diabetic controls emphasizing the time the time-dependent efficacy of turmeric in restoring metabolic homeostasis.<sup>16</sup>

Curcumin, the bioactive compound of turmeric is ascribed for the gradual modification of lipid picture after extended use due to its influence on lipid metabolism through different mechanisms.<sup>17</sup> Another study indicates that the expression of peroxisome proliferator-activated receptor gamma (PPAR- $\gamma$ ) and liver X receptors (LXRs) both essential regulators of cholesterol release and lipoprotein metabolism, are increased in response to curcumin.<sup>18</sup> Helping to lower oxidative stress is curcumin's antioxidant properties. By lowering lipid peroxidation and enhancing the body's natural antioxidant defences, this oxidative stress is vital in the development of dyslipidemia connected to diabetes.<sup>19</sup> By week twelve, the lipid levels are seen to rise in diabetic control group whereas the turmeric treated diabetic group has significant decrease in lipid profile. The findings conform to the study by Muhammed WI, with the difference that dyslipidemia was introduced by intraperitoneal dexamethasone.<sup>20</sup>

These results support earlier studies showing curcumin's ability to lower total cholesterol, triglycerides, and LDL-C while raising HDL-C in diabetic mice.<sup>21</sup> A meta-analysis<sup>22</sup> of preclinical studies stressing the duration-dependent efficacy of phytochemicals in metabolic diseases supports the conclusion that sustained administration is necessary for attaining clinically significant outcomes as observed in the current study. The normalization of lipid markers in the turmeric-treated group matches clinical observations showing better cardiovascular risk profiles in diabetic patients on curcumin supplementation.<sup>23</sup>

Despite these positive results, the study used an animal model which does not fully reflect human metabolic complexity. Future research should look into turmeric's translational potential in human trials, particularly with relation to long-term effects on diabetic dyslipidemia and cardiovascular outcomes. Clinical trials with the use of purified curcumin instead of raw turmeric powder can produce much better effects.

## CONCLUSION

The improvements in lipid profile parameters (total serum cholesterol, phospholipids, and LDL/HDL ratio) are observed in this study with the use of turmeric powder when comparison was made among normal control, diabetic control, and diabetic rats treated with turmeric (curcuminoids).

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### Address for Correspondence:

**Dr Aysa Zaheer**, Associate Professor, Department of Physiology, Services Institute of Medical Sciences, Lahore, Pakistan. **Cell:** +92-333-4222413  
**Email:** ayszahaheer33@gmail.com

**Received:** 12 Jul 2025

**Reviewed:** 26 Dec 2025

**Accepted:** 31 Dec 2025

### Contribution of Authors:

**JL:** Conceptualizing, data collection, literature review, drafting manuscript

**AZ:** Critically reviewing the manuscript

**MJS:** Interpretation of data

**SM:** Statistical analysis

**IQ:** Editing of manuscript

**SN:** Literature review

**Conflict of Interest:** None to declare

**Funding source:** None to declare