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Study of the Impact on Natural Polyphenolic Compounds on liver Pathology Induced by a High-fat Diet Combined with Streptozotocin

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Abstract

Metabolic syndrome-associated fatty liver disease (MAFLD) is a growing global health concern, with its prevalence increasing steadily. Statistical data suggests a dramatic rise from 18.2% in the 1990s to a potential 40% in 2020-2024, with projections exceeding 50% of the world's population within the next decade.

The full spectrum of metabolic disorders contributing to MAFLD pathogenesis remains unclear. Additionally, no specific pharmaceutical treatment currently exists. Since oxidative stress seems to be a key factor, research has focused on the potential therapeutic and preventative role of antioxidant compounds, particularly polyphenols.

Since this pathology is mainly associated with peroxidation, the search for its therapeutic and preventive means is directed towards studying antioxidant compounds, including polyphenols, in relation to the aforementioned pathology.

This study aimed to evaluate the impact of polyphenolic compounds on organ-specific functional markers in animal models with high-fat diet and streptozotocin-induced liver pathology.

Following ethical guidelines, the study employed a multi-stage approach. First, a high-fat diet and streptozotocin were used to establish a pathological model in rats, confirmed by appropriate markers. These animals then received either a polyphenolic fraction extracted from grape seeds (developed by our team) or pure quercetin. Finally, the study assessed the antioxidant properties of these compounds and their impact on plasma levels of biochemical markers (ALT, AST, cholesterol, and triglycerides).

In conclusion, this study suggests that polyphenolic compounds, particularly quercetin, have a beneficial effect on liver function markers (ALT/AST) in a high-fat diet and streptozotocin-induced rat model of MAFLD. While further research is needed to explore their long-term effects on cholesterol and triglyceride metabolism (a 10-day course did not show a significant impact on cholesterol and triglyceride metabolism), these findings offer promising possibilities for the development of novel therapeutic strategies for MAFLD.

Study Results and Their Discussion:

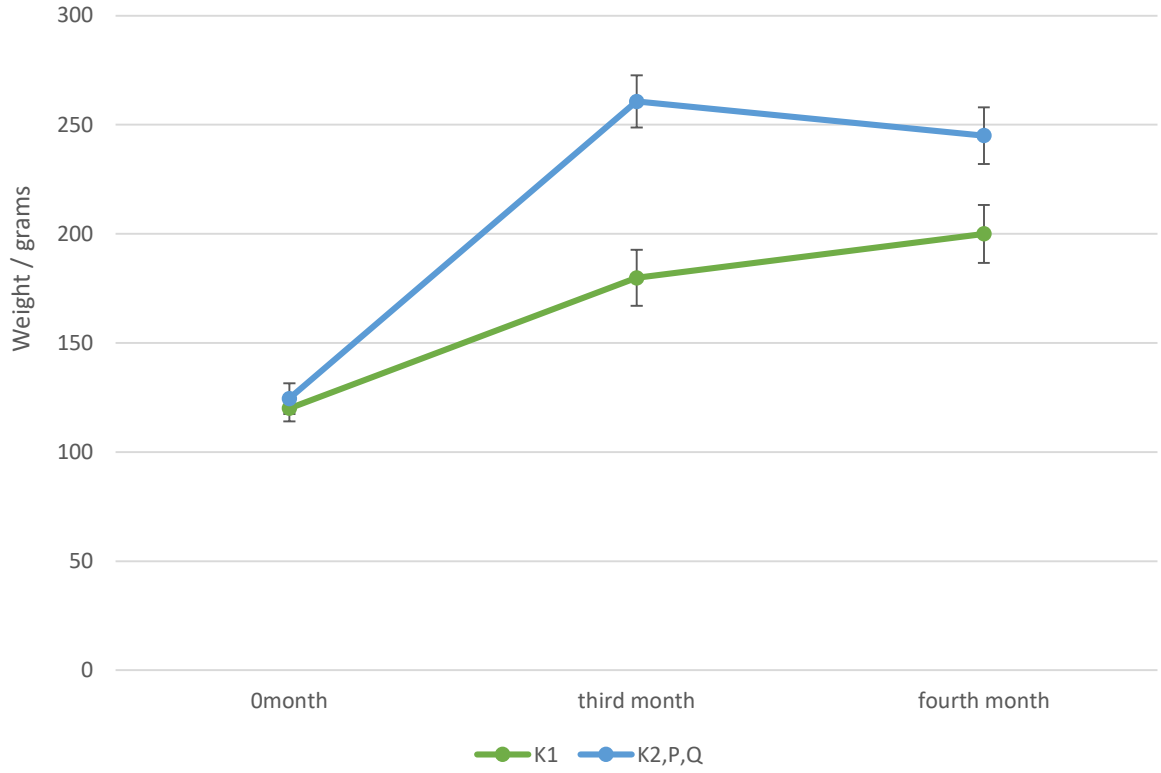

Figure 1 Weight change in study groups,
K1 - Standard diet; K2 - High-fat diet

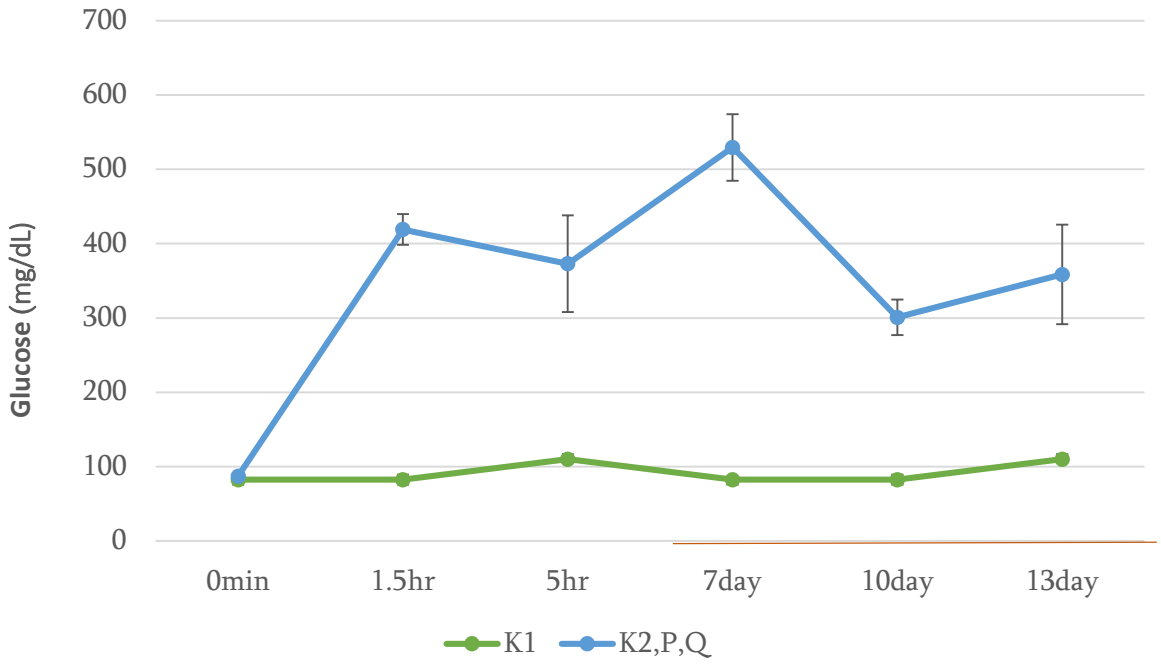

Figure 2 Changes in blood glucose concentration in animals after streptozotocin injection
K1 - Standard diet; K2, P, Q - High-fat diet

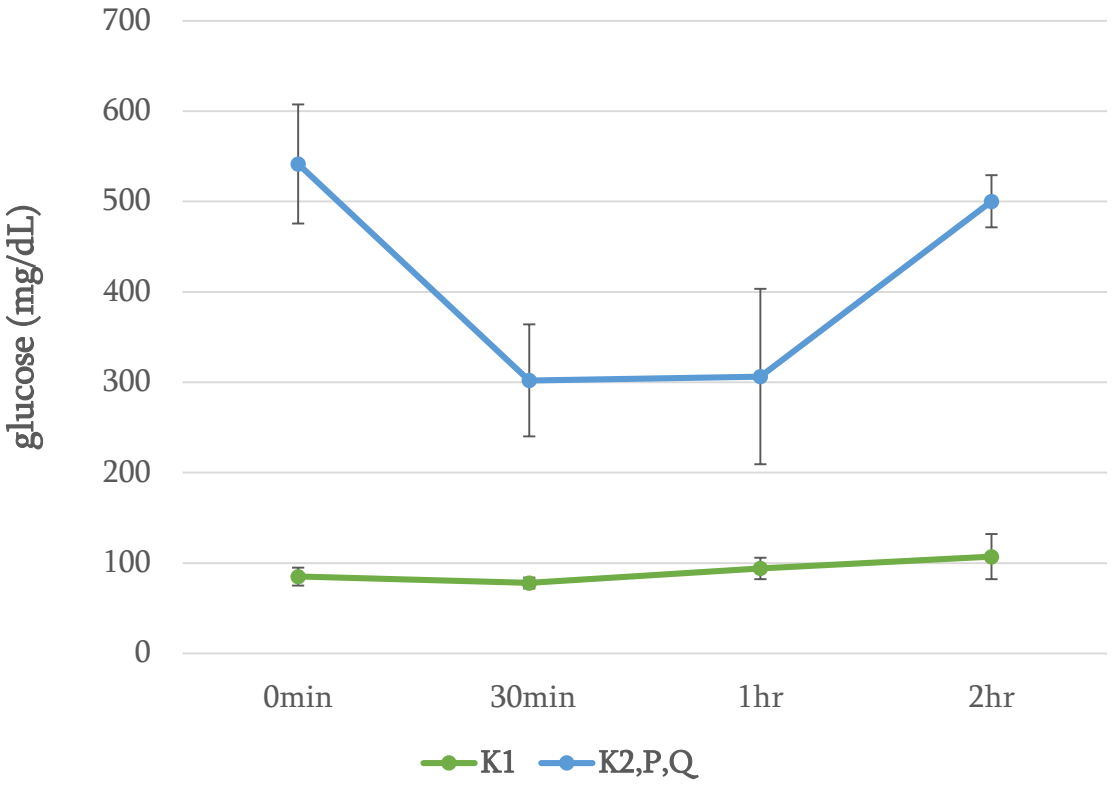

Figure 3 Changes in blood glucose concentration in animals after insulin injection
K1 - Standard diet; K2, P, Q - High-fat diet

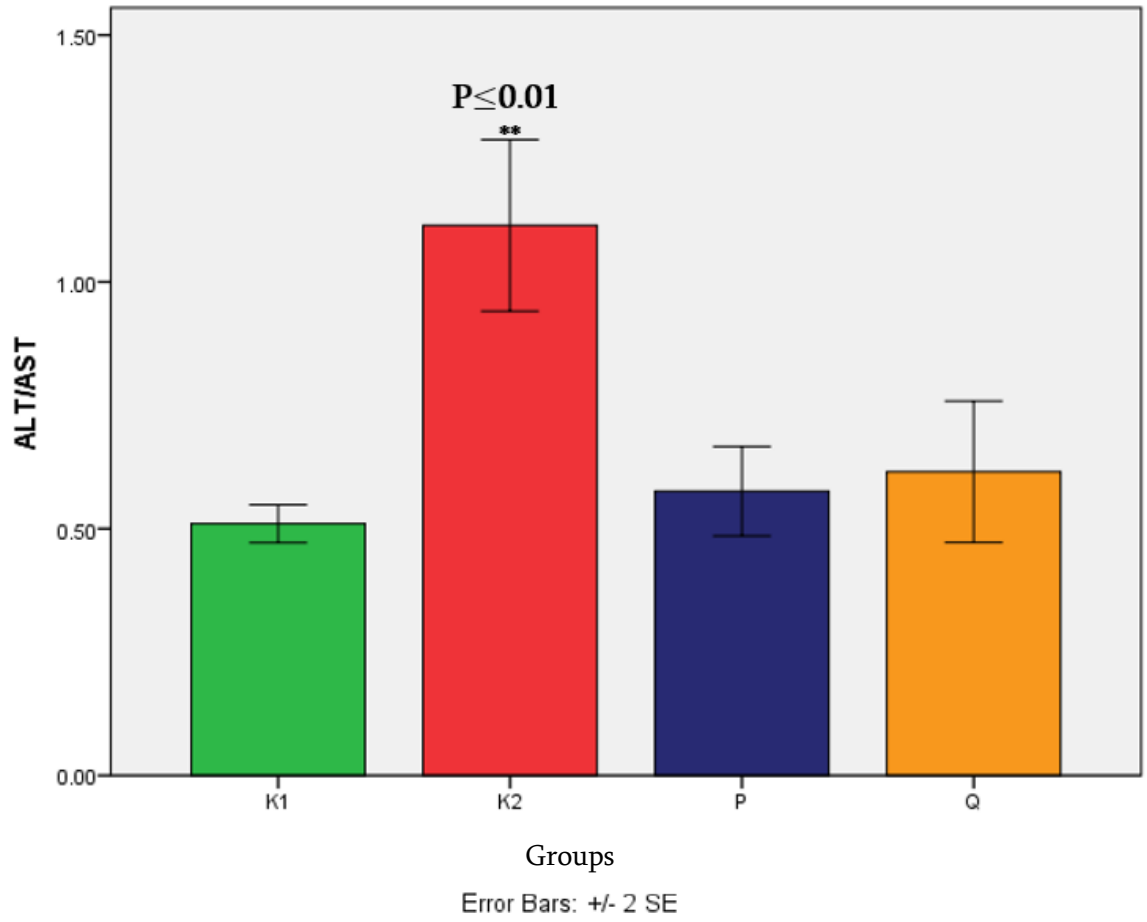

Figure 4 Changes in ALT/AST ratio between healthy and pathological groups

I. A control group (healthy)
II. A pathological group (Pathological liver condition induced by streptozotocin)
III. A group treated with polyphenols
IV. A group treated with quercetin

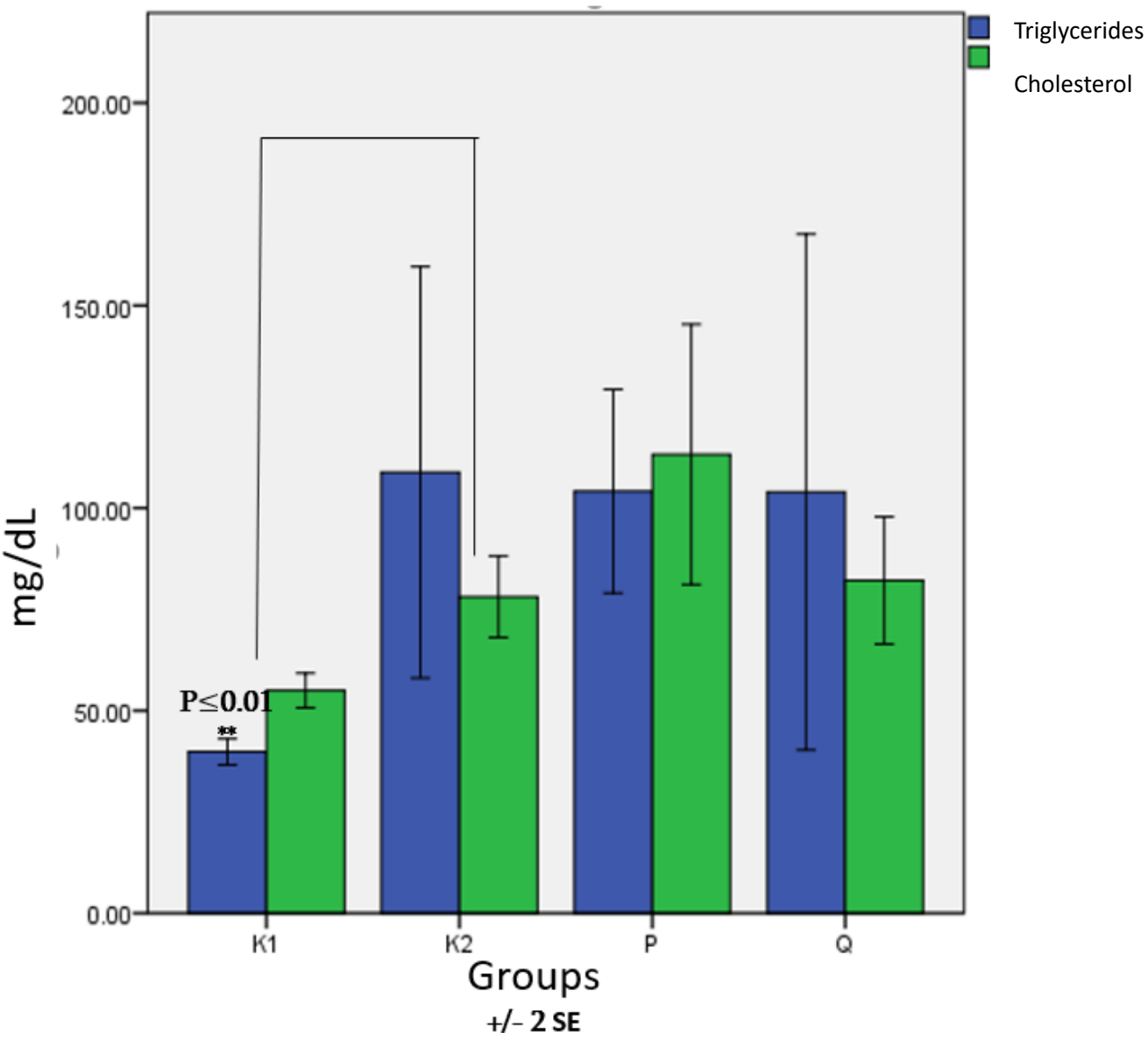

Figure 5 Quantitative indicators of cholesterol and triglycerides

I. A control group (healthy)
II. A pathological group (Pathological liver condition induced by streptozotocin)
III. A group treated with polyphenols
IV. A group treated with quercetin

Table 1. One-way analysis of variance

ANOVA

Ratio

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	1.892	3	.631	27.543	.000
Within Groups	.847	37	.023		
Total	2.739	40			

Table 2. Multiple comparisons between groups

Multiple Comparisons

Dependent Variable: Ratio

	(I) Groups	(J) Groups	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
Tukey HSD	K1	K2	-.60434*	.06857	.000	-.7888	-.4199
		P	-.06579	.05927	.686	-.2252	.0936
		Q	-.10519	.06857	.428	-.2896	.0792
	K2	K1	.60434*	.06857	.000	.4199	.7888
		P	.53856*	.07316	.000	.3418	.7353
		Q	.49915*	.08088	.000	.2816	.7167
	P	K1	.06579	.05927	.686	-.0936	.2252
		K2	-.53856*	.07316	.000	-.7353	-.3418
		Q	-.03940	.07316	.949	-.2362	.1574
	Q	K1	.10519	.06857	.428	-.0792	.2896
		K2	-.49915*	.08088	.000	-.7167	-.2816
		P	.03940	.07316	.949	-.1574	.2362
Bonferroni	K1	K2	-.60434*	.06857	.000	-.7955	-.4132
		P	-.06579	.05927	1.000	-.2310	.0994
		Q	-.10519	.06857	.801	-.2963	.0860
	K2	K1	.60434*	.06857	.000	.4132	.7955
		P	.53856*	.07316	.000	.3346	.7425
		Q	.49915*	.08088	.000	.2737	.7246
	P	K1	.06579	.05927	1.000	-.0994	.2310
		K2	-.53856*	.07316	.000	-.7425	-.3346
		Q	-.03940	.07316	1.000	-.2433	.1645
	Q	K1	.10519	.06857	.801	-.0860	.2963
		K2	-.49915*	.08088	.000	-.7246	-.2737
		P	.03940	.07316	1.000	-.1645	.2433

*. The mean difference is significant at the 0.05 level.

We can summarize the obtained results as follows:

The different diets among the groups over three months revealed significant results. In the experimental group of animals fed a high-fat diet, their weight increased much more compared to the control group, which received a standard diet (Fig. 1). These results indicate that the high-fat diet played a significant role in increasing the body mass of the experimental animals.

Excess weight is considered one of the main risk factors for the development of non-alcoholic fatty liver disease. Accordingly, our experimental model, which caused weight gain in animals in response to high-fat feeding, could be used to study this disease and search for potential treatment methods.

To assess the degree of hyperglycemia induced by streptozotocin, the change in glucose concentration (mg/dL) in the blood of each animal was determined dynamically. Blood glucose content was measured using a glucometer at different times after both doses of streptozotocin injection in both control and experimental group animals.

Figure 2 shows the change in blood glucose concentration in adult rats during the first 48 hours after streptozotocin injection. The figure shows that 1.5 hours after streptozotocin injection, the amount of glucose in the blood of the experimental group rats increased approximately five-fold compared to the control animals. Over the next 3.5 hours (1.5-5 hours), 95% of the experimental animals showed a tendency for blood glucose concentration to decrease. Twenty-four hours after injection, blood glucose concentration increased again. Elevated glucose levels were maintained until the end of the experiment, indicating that stable hyperglycemia was achieved after the second dose of streptozotocin injection.

A similar trend was observed as a result of insulin injection (Fig. 3): thirty minutes after injection, a decrease in blood glucose levels was observed, but this effect was not long-lasting. One hour after injection, blood glucose levels began to increase, and after two hours, hyperglycemia persisted. This indicates that β cells retain the ability to synthesize and secrete insulin, which is consistent with literature data suggesting that a low dose of streptozotocin injection does not destroy the entire population of pancreatic β cells. Accordingly, at this concentration, glucose does not lose sensitivity to exogenous insulin, although hyperglycemia persists in the blood.

Regarding liver-specific characteristics, the increase in the ALT/AST ratio in group K2 indicates a disruption of liver function caused by a fatty diet and streptozotocin injections. For the third (P) and fourth (Q) groups, Fig. 4 shows that in the polyphenol-treated group (P), compared to group K2, the ALT/AST ratio decreases, indicating an improvement in the pathological condition. A similar trend is observed in the quercetin-treated group (Q). Thus, we can conclude that during a 10-day series of injections, polyphenols and quercetin exhibit a hepatoprotective effect by reducing the ALT/AST ratio in plasma, although they do not affect cholesterol and triglyceride metabolism (Fig. 5).