International Journal of Advanced Statistics and Probability, 12 (2) (2025) 1-8

International Journal of Advanced Statistics and Probability

Website: www.sciencepubco.com/index.php/IJASP
https://doi.org/10.14419/rd686p43
Research paper

A 2^k Factorial Design on The Effect of Different Cholesterol Reducing Factors

Ukeme P. Akra [1] *, Anthony A. Isaac [1], Edet E. Bassey [2], Andrew C. Etim [1], Usenobong B. Akpan [3], Itoro T. Michael [1]

[1] Department of Statistics, Akwa Ibom State University, Ikot Akpaden
[2] Department of Statistics, University of Calabar, Calabar
[3] Department of Physics, Akwa Ibom State University, Ikot Akpaden
**Corresponding author E-mail: ukemeakra@aksu.edu.ng*

Received: May 14, 2025, Accepted: June 5, 2025, Published: June 11, 2025

Abstract

This research focuses on the application of a 2^K factorial design on the effect of different cholesterol-reducing factors. The purpose is to determine the effect of each factor and its interactions that contribute to the reduction of cholesterol levels. A complete 2^4 factorial design with two replicates was used for the experiment, where the factors are: Frequency (A), Duration (B), Dosage (C) and Drug type (D) each at two levels. The levels are A (Once and Twice daily), B (4 and 8 weeks), C (Low and High), and D (Statins and PCKS9 inhibitor). The analysis of variance technique was used to test the significant effects, and a Pareto chart was plotted to show a particular factor that has a higher reduction strength to reduce cholesterol level. Results revealed that all the main effects are significant, two–factor interactions of six two–factor interactions are not significant, three–factor interactions are not significant, and four–factor interactions is significant at the 5% level of significance. Hence, from the Pareto chart, it is confirmed that factor C outperformed than other factors and their interactions in reducing cholesterol level.

Keywords: *Cholesterol; Reduction; Factor; Level; Factorial Design.*

1. Introduction

Medications that lower cholesterol are a cornerstone in the treatment of elevated cholesterol, a major contributor to cardiovascular conditions such as heart attacks and strokes. Among these medications, statins are considered the first-line therapy due to their well-established ability to lower LDL cholesterol and reduce the incidence of serious cardiovascular events, including mortality. For individuals who cannot tolerate statins or require additional lipid-lowering therapy, alternatives such as PCSK9 inhibitors, ezetimibe, fibrates, bile acid sequestrants, and niacin are viable options. Treatment strategies should be customized according to each patient's lipid profile, accompanying medical conditions, and cardiovascular risk level. Pharmacologic interventions should be complemented by lifestyle adjustments such as healthy eating, regular physical activity, and weight control. Medical guidance is essential to determine the most suitable approach to cholesterol and cardiovascular risk management. Statins also possess additional properties beyond lipid-lowering known as pleiotropic effects including anti-inflammatory action (evidenced by reduced CRP levels), antioxidant activity, anti-thrombotic effects, inhibition of cellular proliferation, improvement of endothelial function, and mitigation of vascular remodeling. Although these effects are consistently observed, their exact role in statins' overall cardiovascular benefit remains unclear [1].

Differences in statins' pharmacokinetic characteristics may lead to variation in their safety profiles and drug interactions. Most statins are lipophilic, allowing easier cellular penetration, except for pravastatin and rosuvastatin, which are hydrophilic. The clinical implications of this distinction are not fully defined. In patients with statin intolerance, dosing strategies such as alternate-day or biweekly administration of long-acting statins have been explored. While short-acting statins are most effective when taken at night, aligned with the circadian rhythm of HMG-CoA reductase activity, long-acting statins demonstrate comparable efficacy regardless of administration time [2 - 6]. The SAMSON trial offered a significant insight into statin intolerance, revealing that up to 90% of side effects attributed to statins were also observed with placebos, underscoring the nocebo effect. Notably, half of the participants were able to restart statin therapy successfully [7]. Despite this, perceived intolerance remains a common cause for discontinuation and contributes to suboptimal LDL-C management [8].

In individuals without diabetes risk factors, statins generally do not increase diabetes risk. Although FDA labeling includes a warning about potential cognitive side effects, randomized trials have not substantiated a meaningful connection [9 - 11]. While a minor elevation in stroke risk has been suggested in secondary prevention cases, the overall benefit in reducing vascular events outweighs this slight risk [12]. Liver enzyme elevations associated with statin use are typically mild, dose-dependent, and rarely serious. Baseline liver function testing is recommended before starting therapy, but routine monitoring is no longer deemed necessary. The incidence of statin-induced liver failure

mirrors that of the general population, making it an exceedingly rare event [13]. Nonetheless, active liver disease and pregnancy are contraindications to statin use, and women of reproductive age should discuss contraception when using statins [14]. PCSK9 is a liver-produced protease that degrades LDL receptors, thereby influencing LDL cholesterol levels. Inhibiting PCSK9 has proven effective in lowering LDL-C [15 - 17]. PCSK9 inhibitors have demonstrated safety with minimal side effects, mostly limited to injection-site reactions and mild upper respiratory symptoms. No significant evidence has emerged linking them to liver toxicity, muscle issues, cognitive impairment, or drug interactions [18 - 19]. Clinical trials such as the HUYGENS study have shown that combining statins with evolocumab leads to favorable modifications in coronary artery plaque, promoting stabilization and regression [20]. Similarly, the PACMAN-AMI trial reported that adding alirocumab to high-dose rosuvastatin soon after myocardial infarction enhanced plaque reduction in arteries not directly involved in the event [21]. These beneficial effects have also been observed in carotid artery plaques [22 - 23]. Despite their efficacy, the high cost of PCSK9 inhibitors remains a significant barrier to widespread use [24].

Factorial design is a powerful experimental methodology widely used in pharmacological and medical research to evaluate the effects of multiple factors simultaneously. It allows researchers to assess not only the individual (main) effects of treatments or interventions but also the interactions between them. This design is particularly useful in complex biomedical systems where multiple variables can influence outcomes. The application of factorial design is not limited to pharmacological and medical research but can also be applied in other areas like retail business plans and agricultural experiments [25 - 27]. Factorial designs are widely employed in cholesterol-related research, particularly to evaluate the combined effects of lipid-lowering therapies. For instance, [28] utilized a 2×2 factorial randomized controlled trial (RCT) to examine the joint impact of statins and ezetimibe in managing hypercholesterolemia. The study concluded that, although both agents were effective individually, their combined use yielded a synergistic reduction in LDL-C levels. Similarly, [29] applied a 3×2 factorial approach to assess varying doses of atorvastatin, with or without niacin, and found significant interactions between dosage and niacin administration, indicating enhanced reductions in total cholesterol and triglycerides. Functional food interventions have also been explored through factorial designs. A 2×3 factorial study by [30] assessed the effects of plant sterols and dietary fiber on lipid profiles, reporting additive benefits on LDL-C when the two were combined. [31] examined the synergistic effects of olive oil polyphenols and fish oil via a 2×2 factorial design, observing that their combination was more effective in increasing HDL-C and decreasing LDL-C than either component alone. Additionally, factorial trials have been instrumental in evaluating integrated lifestyle and pharmacological strategies. [32] studied the interaction of a low-saturated-fat diet with statin therapy using a 2×2 design, concluding that their combination yielded superior LDL-C reduction. In another study, [33] employed a 3×2 factorial design to explore how varying exercise intensity interacts with statin dosage, revealing that higher-intensity exercise enhances the cholesterol-lowering effects of statins. The versatility of factorial designs also extends to personalized medicine. [34] Implemented a genotype-stratified 2×2 design to assess how APOE genotypes respond to dietary cholesterol and statin therapy, with findings suggesting that individuals with the APOE4 genotype exhibited greater responsiveness to dietary intervention. In preclinical studies, [35] used a 3×3 factorial design in mice to explore the interactions between plant-based compounds and dietary fat, uncovering significant interactions that influenced serum cholesterol and hepatic LDL receptor expression. Finally, factorial designs contribute valuable evidence for public health decision-making. For example, [36] employed such a design to evaluate the combined effects of a mass media campaign and subsidized statin availability in rural communities, emphasizing the policy-relevant utility of factorial trials. The objective of this study is to assess the individual and synergistic effects of various cholesterol-lowering strategies using a factorial design to determine the optimal approach for reducing cholesterol.

2. Methodology

This section describes the techniques used in analyzing the entire work to achieve the aim and objectives of this research.

2.1. Data structure

This study adopted a secondary source of data collection. We collected from the health records unit, General Hospital, Mary Slessor, Calabar. The experiment was carried out using 2^4 factorial design at two levels of drug type (Stanins, PCSK9 inhibitor), dosage (low, high), duration (4 weeks, 8 weeks), and frequency (once, twice daily) with two replications in the adoption of a randomized complete block design. The results (cholesterol reduction level) were obtained, and the layout design structure is shown in Table 1.

Table 1: Layout Design Structure for 2^4 Factorial Design

Trial	Frequency	Duration	Dosage	Drug type	Cholesterol reduction (%)	
1	Once daily	4weeks	Low	Statin	17.1	19.1
2	Twice daily	4weeks	Low	Statin	14.2	14.8
3	Once daily	8weeks	Low	Statin	13.5	15.3
4	Twice daily	8weeks	Low	Statin	16.7	15.5
5	Once daily	4weeks	High	Statin	12.3	13.8
6	Twice daily	4weeks	High	Statin	12.5	12.6
7	Once daily	8weeks	High	Statin	14.6	14.2
8	Twice daily	8weeks	High	Statin	12.9	12.7
9	Once daily	4weeks	Low	PCSK 9	20.4	21.9
10	Twice daily	4weeks	Low	PCSK 9	18.6	18.5
11	Once daily	8weeks	Low	PCSK 9	17.9	19.5
12	Twice daily	8weeks	Low	PCSK 9	14.2	15.9
13	Once daily	4weeks	High	PCSK 9	18.1	19.2
14	Twice daily	4weeks	High	PCSK 9	13.4	12.9
15	Once daily	8weeks	High	PCSK 9	14.6	15.3
16	Twice daily	8weeks	High	PCSK 9	13.8	13.5

2.2. Estimation of effects

In this research, four factors, A (Frequency), B (Duration), C (Dosage), and D (Drug type) are of interest, each at two levels. The design is 2^4 factorial design with sixteen (16) treatment combinations ,and the standard order for the treatment combination is written as;:(1) a, b, ab, c, ac, bc, abc, d, ad, bd, abd, cd, acd, bcd, abcd. The effects in 2^4 factorial design can be obtained in an algebraic form shown in Table 2 and analysis of variance format is displayed in Table 3.

Table 2: Algebraic Signs for Calculating Effects in the 2^4 Design

T.C	A	B	AB	C	AC	BC	ABC	D	AD	BD	ABD	CD	ACD	BCD	ABCD
(1)	-	-	+	-	+	+	-	-	+	+	-	+	-	-	+
a	+	-	-	-	-	+	+	-	-	+	+	+	+	-	-
b	-	+	-	-	+	-	+	-	+	-	+	+	-	+	-
ab	+	+	+	-	-	-	-	-	-	-	-	+	+	+	+
c	-	-	+	+	-	-	+	-	+	+	-	-	+	+	-
ac	+	-	-	+	+	-	-	-	-	+	+	-	-	+	+
bc	-	+	-	+	-	+	-	-	+	-	+	-	+	-	+
abc	+	+	+	+	+	+	+	-	-	-	-	-	-	-	-
d	-	-	+	-	+	+	-	+	-	-	+	-	+	+	-
ad	+	-	-	-	-	+	+	+	+	-	-	-	-	+	+
bd	-	+	-	-	+	-	+	+	-	+	-	-	+	-	+
abd	+	+	+	-	-	-	-	+	+	+	+	-	-	-	-
cd	-	-	+	+	-	-	+	+	-	-	+	+	-	-	+
acd	+	-	-	+	+	-	-	+	+	-	-	+	+	-	-
bcd	-	+	-	+	-	+	-	+	-	+	-	+	-	+	-
abcd	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

The sum of squares for the main effects and all the interactions are obtained from Table 2.

$$SS_A = \frac{\left[\begin{matrix} a+ab+ac+abc+ad+abd+acd+abcd-(1) \\ -a-c-bc-d-db-cd-bcd \end{matrix}\right]^2}{16n} \tag{1}$$

$$SS_B = \frac{\left[\begin{matrix} b+ab+bc+abc+bd+abd+bcd+abcd-(1) \\ -a-c-ac-d-ad-cd-acd \end{matrix}\right]^2}{16n} \tag{2}$$

$$SS_C = \frac{\left[\begin{matrix} c+ac+bc+abc+cd+acd+bcd+abcd-(1) \\ -a-b-ab-d-ad-bd-abd \end{matrix}\right]^2}{16n} \tag{3}$$

$$SS_D = \frac{\left[\begin{matrix} d+ad+bd+abd+cd+acd+bcd+abcd-(1) \\ -a-b-ab-c-ac-bc-abc \end{matrix}\right]^2}{16n} \tag{4}$$

$$SS_{AB} = \frac{\left[\begin{matrix} abcd-bcd-acd+cd+abd-bd-ad+d+abc \\ -bc-ac+c+ab-b-a-(1) \end{matrix}\right]^2}{16n} \tag{5}$$

$$SS_{AC} = \frac{\left[\begin{matrix} (1)-a+b-ab-a+ac-bc+abc+d-ad+bd \\ -abd-cd+acd-bcd+abcd \end{matrix}\right]^2}{16n} \tag{6}$$

$$SS_{AD} = \frac{\left[\begin{matrix} (1)-a+b-ab+c-ac+bc-abc-d+ad-bd \\ +abd-cd+acd-bcd+abcd \end{matrix}\right]^2}{16n} \tag{7}$$

$$SS_{BC} = \frac{\left[\begin{matrix} (1)+a-b-ab-c-ac+bc+abc+d+ad-bd \\ -abd-cd-acd+bc+abcd \end{matrix}\right]^2}{16n} \tag{8}$$

$$SS_{BD} = \frac{\left[\begin{matrix} (1)+a-b-ab+c+ac-bc-abc-d-ad+bd \\ +abd-cd-acd+bcd+abcd \end{matrix}\right]^2}{16n} \tag{9}$$

$$SS_{CD} = \frac{\left[\begin{matrix} (1)+a+b+ab-c-ac-bc-abc-d-ad-bd \\ -abd+cd+acd+bcd+abcd \end{matrix}\right]^2}{16n} \tag{10}$$

$$SS_{ABC} = \frac{\left[\begin{matrix} abcd-bcd-acd+cd-abd+bd+ad-d+abc \\ -bc-ac+c-ab+b+a-(1) \end{matrix}\right]^2}{16n} \tag{11}$$

$$SS_{ABD} = \frac{\left[\begin{matrix} abcd - bcd - acd + cd + abd - bd - ad + d - abc \\ + bc + ac - c - ab + b + a - (1) \end{matrix}\right]^2}{16n} \tag{12}$$

$$SS_{ACD} = \frac{\left[\begin{matrix} abcd - bcd + acd - cd - abd + bd - ad + d - abc \\ - bc + ac + c + ab + b - a - (1) \end{matrix}\right]^2}{16n} \tag{13}$$

$$SS_{BCD} = \frac{\left[\begin{matrix} abcd + bcd - acd - cd - abd - bd + ad + d - abc \\ - bc + ac + c + ab + b - a - (1) \end{matrix}\right]^2}{16n} \tag{14}$$

$$SS_{ABCD} = \frac{\left[\begin{matrix} (1) - a - b + ab - c + ac + bc - abc - d + ad + bd \\ - abd + cd - acd - bcd + abcd \end{matrix}\right]^2}{16n} \tag{15}$$

$$SS_{Total} = \sum_{i}^{a}\sum_{j}^{b}\sum_{k}^{c}\sum_{l}^{d}\sum_{m}^{n} Y_{ijklm}^{2} - \frac{Y_{....}^{2}}{N} \tag{16}$$

SS_{Total} = Subtracting all the effects from SS_{Total}

Table 3: Structural form of Analysis of Variance Table

Source of Variation	SS	Df	MSS	F-Value
Main effects				
A	SS_A	1	MS_A	F_A
B	SS_B	1	MS_B	F_B
C	SS_C	1	MS_C	F_C
D	SS_D	1	MS_D	F_D
Two-factor interactions				
AB	SS_{AB}	1	MS_{AB}	
AC	SS_{AC}	1	MS_{AC}	F_{AB}
AD	SS_{AD}	1	MS_{AD}	F_{AC}
BC	SS_{BC}	1	MS_{BC}	F_{AD}
BD	SS_{BD}	1	MS_{BD}	F_{BC}
CD	SS_{CD}	1	MS_{CD}	F_{BD}
Three-factor interactions				F_{CD}
ABC	SS_{ABC}	1	MS_{ABC}	
ABD	SS_{ABD}	1	MS_{ABD}	F_{ABC}
ACD|	SS_{ACD}	1	MS_{ACD}	F_{ABD}
BCD	SS_{BCD}	1	MS_{BCD}	F_{ACD}
Four-factor interactions				F_{BCD}
ABCD	SS_{ABCD}	1	MS_{ABCD}	
Error	SS_E	$2^k(n-1)$	MS_E	F_{ABCD}
Total	SS_T	$n2^k - 1$		

2.3 Assumptions of ANOVA

1) Normality: The data's normal distribution was evaluated using a Q–Q (quantile–quantile) plot. This graphical method compares observed data quantiles with theoretical quantiles from a normal distribution. A linear pattern in the plot suggests approximate normality.
2) Homoscedasticity (Equal Variances): Bartlett's test was used to assess whether variances across groups are equal. This test relies on a likelihood ratio and assumes the test statistic follows a Chi-square distribution with k – 1 degrees of freedom, where k is the number of groups.

$$\chi^2 = \frac{(N-1)\ln S_p^2 - \sum_{i=1}^{k}(n_i - 1)\ln S_i^2}{1 + \frac{1}{3(k-1)}\left(\sum_{i=1}^{k}\frac{1}{n_i - 1} - \frac{1}{N-k}\right)} \tag{17}$$

Where k is the number of groups, n_i is the sample size in the group $_i$, S_i^2 is the sample variance of the group $_i$, $N = \sum_{i=1}^{k} n_i$ is the total number of observations, and S_p^2 is pooled variance, defined as; $S_p^2 = \frac{1}{n-k}\sum_{i=1}^{k}(n_i - 1)S_i^2$. The decision rule states that if $P < C$ (i.e 0.05), H_0 is rejected and variances are not equal. Otherwise, failed to reject H_0 and conclude that variances are assumed equal.

3. Results

Building on the experimental design, the following results are obtained.

3.1. Test of assumptions of ANOVA

i) Normality test. Figure 3.1 shows that the points are closely aligned with the diagonal reference, suggesting that data is approximately normally distributed.

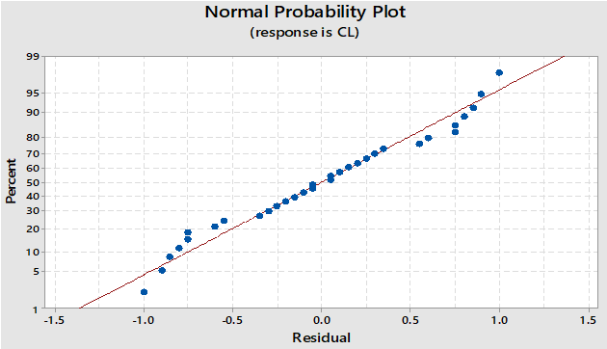

Figure 3.1: Normality Plot Showing the Distribution of the Data.

ii) Homoscedasticity test (Test for Equal Variances)

Method	Test statistic	P – value
Bartlett	12.99	0.603

The P - P-value is greater than 0.05 (i.e., 0.603>0.05), suggesting that the assumption of equal variance (homoscedasticity) is not violated. Therefore, an ANOVA is appropriate for the experiment.

3.2. Analysis of main effects and interactions

The main effect, two–factor interactions, three–factor effect interactions, and four–factor interactions are shown in Table 4, the model summary in Table 5, and the test of coefficients of the terms in Table 6. Pareto chart demonstrated that the true picture of the results is also obtained in this section (see Figure 2).

Table 4: Analysis of Variance for the 2^4 Factorial Design of the Studied Experiment

Source	DF	Adj SS	Adj MS	F-Value	P-Value
Model	15	206.362	13.7575	20.72	0.000
Linear	4	156.406	39.1016	58.88	0.000
A	1	36.338	36.3378	54.72	0.000
B	1	11.640	11.6403	17.53	0.001
C	1	68.153	68.1528	102.63	0.000
D	1	40.275	40.2753	60.65	0.000
2-Way Interactions	6	32.559	5.4266	8.17	0.000
A*B	1	6.753	6.7528	10.17	0.006
A*C	1	0.070	0.0703	0.11	0.749
A*D	1	10.238	10.2378	15.42	0.001
B*C	1	5.200	5.2003	7.83	0.013
B*D	1	9.353	9.3528	14.08	0.002
C*D	1	0.945	0.9453	1.42	0.250
3-Way Interactions	4	0.431	0.1078	0.16	0.954
A*B*C	1	0.165	0.1653	0.25	0.625
A*B*D	1	0.138	0.1378	0.21	0.655
A*C*D	1	0.015	0.0153	0.02	0.881
B*C*D	1	0.113	0.1128	0.17	0.686
4-Way Interactions	1	16.965	16.9653	25.55	0.000
A*B*C*D	1	16.965	16.9653	25.55	0.000
Error	16	10.625	0.6641		
Total	31	216.987			

Table 5: Model Summary

S	R-sq	R-sq(adj)	R-sq(pred)
0.814900	95.10%	90.51%	80.41%

Table 6: Test of Coefficients of the Terms in the Model

Term	Effect	Coef	SE Coef	T-Value	P-Value	VIF
Constant		15.609	0.144	108.36	0.000	
A	-2.131	-1.066	0.144	-7.40	0.000	1.00
B	-1.206	-0.603	0.144	-4.19	0.001	1.00
C	-2.919	-1.459	0.144	-10.13	0.000	1.00
D	2.244	1.122	0.144	7.79	0.000	1.00
A*B	0.919	0.459	0.144	3.19	0.006	1.00

A*C	-0.094	-0.047	0.144	-0.33	0.749	1.00
A*D	-1.131	-0.566	0.144	-3.93	0.001	1.00
B*C	0.806	0.403	0.144	2.80	0.013	1.00
B*D	-1.081	-0.541	0.144	-3.75	0.002	1.00
C*D	-0.344	-0.172	0.144	-1.19	0.250	1.00
A*B*C	-0.144	-0.072	0.144	-0.50	0.625	1.00
A*B*D	-0.131	-0.066	0.144	-0.46	0.655	1.00
A*C*D	-0.044	-0.022	0.144	-0.15	0.881	1.00
B*C*D	-0.119	-0.059	0.144	-0.41	0.686	1.00
A*B*C*D	1.456	0.728	0.144	5.05	0.000	1.00

The fitted equation model is given as;

CL = 15.609 - 1.066 A - 0.603 B - 1.459 C + 1.122 D + 0.459 A*B - 0.047 A*C - 0.566 A*D + 0.403 B*C - 0.541 B*D - 0.172 C*D - 0.072 A*B*C - 0.066 A*B*D - 0.022 A*C*D - 0.059 B*C*D + 0.728 A*B*C*D

3.3. Pareto chart

The Pareto chart identified dosage (C) as the most influential factor, displaying the highest standardized effect among all variables. This suggests that dosage is not only statistically significant but also practically impactful in reducing cholesterol levels. Despite the statistical significance of all main effects (A, B, C, D), dosage exhibited the greatest magnitude of effect, highlighting its importance in treatment planning. While certain two-factor interactions involving dosage (e.g., A×C and C×D) were not significant, the dosage's effect remained consistently strong across different drug types and dosing frequencies. The bars (effects) above the reference line are significant, which shows how different effects and interactions contribute to the cholesterol reduction level in patients. The significance of dosage (D) as a dominant factor implies that changes from low to high levels of dosage cause a statistically significant difference in cholesterol reduction, independent of other factors. Regardless of which drug is used, how long it's taken, or how often it's administered, the dosage level by itself has a strong, measurable effect on cholesterol levels. This suggests that optimizing dosage has the biggest potential for improving patient outcomes.

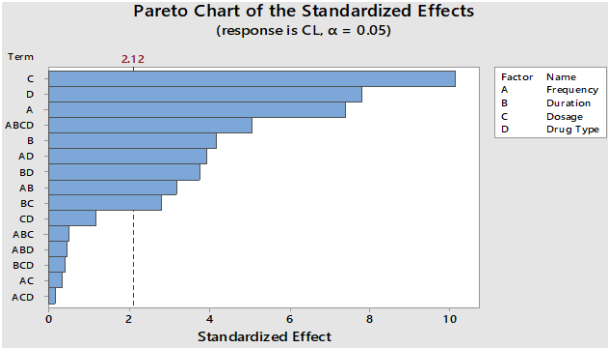

Fig. 2: Pareto Chart for the Factor Effect.

4. Discussion of results

The results in Table 4 revealed that all the main effects, frequency, treatment duration, drug type, and dosage, were found to significantly influence cholesterol levels. Several two-way interactions (B×C, A×D, B×D, A×B) were also statistically significant, while A×C and C×D were not. This suggests that the interaction between certain factor pairs, such as dosage and duration, has a meaningful effect, whereas others, like dosage with frequency or drug type, do not. No significant three-way interactions were observed, indicating that tri-factor combinations do not produce additional effects beyond those explained by two-way interactions. However, the four-way interaction (A×B×C×D) was significant at the 5% level, implying that under specific combinations of all four factors, cholesterol levels may respond in an unpredictable or non-linear manner due to synergistic or antagonistic influences. In Table 5, the model's coefficient of determination (r) indicates a strong explanatory power, with over 95% of the variation in cholesterol reduction explained by the factors in the model. This high value reflects an excellent model fit. Table 6 results confirmed that each factor, drug type, frequency, duration, and dosage, significantly contributes to cholesterol reduction. Specifically, the B×D interaction (Duration × Drug Type) suggests that drug efficacy may vary depending on the length of treatment. The A×C interaction (Frequency × Dosage) implies that treatment effectiveness at various dosages is influenced by how often the drug is administered. The B×C interaction (Duration × Dosage) indicates that a longer treatment duration may be required to fully realize the benefits of higher dosages.

On the other hand, the non-significance of A×C and C×D interactions indicates that dosage effects are consistent regardless of frequency and drug type, suggesting a uniform dosing strategy could be applied across these variables. The presence of a significant four-way interaction suggests complex, higher-order dependencies that require further study. This finding aligns with prior research: [37] observed that higher statin doses produced greater LDL-C reductions irrespective of drug type. [38] Reported similar findings for rosuvastatin, where dose escalation yielded more substantial effects than frequency or type changes. [39] Concluded in a meta-analysis that dosage is the most reliable determinant of cholesterol reduction, while frequency and duration have more variable impacts. [40] Also observed dosage as the strongest main effect in a factorial trial involving antihypertensive medications.

5. Conclusion

The analysis reveals that all four investigated factors, frequency, duration, dosage, and drug type, independently and significantly affect cholesterol reduction. Careful adjustment of each parameter is crucial for optimizing treatment outcomes. Significant two-way interactions

(such as B×C, A×D, A×B) reinforce the importance of considering the interdependence between treatment variables when designing therapeutic regimens. Although no three-way interactions were statistically significant, the presence of a significant four-way interaction suggests that complex interdependencies may exist when all factors interact simultaneously. This warrants cautious interpretation and further investigation. Among all variables, dosage emerged as the most impactful factor both statistically and practically. Therefore, treatment strategies should emphasize accurate dosage calibration to achieve optimal cholesterol-lowering effects.

Recommendations

i) Clinical protocols should prioritize dosage selection for improved efficacy.
ii) Future studies should explore expanded dosage levels or adopt composite experimental designs to fine-tune dosage strategies.
iii) Additional research should consider integrating lifestyle modifications to better understand combined intervention effects.

References

[1] Joshi, P. H., & Jacobson, T. A. (2010). Therapeutic options to further lower C-reactive protein for patients on statin treatment. *Curr Atheroscler Rep.* 12:34-42 https://doi.org/10.1007/s11883-009-0075-x.
[2] Causevic-Ramosevac, A., & Semiz, S. (2013). Drug interactions with statins. *Acta Pharm.* 63:277-293https://doi.org/10.2478/acph-2013-0022.
[3] Hu, M., & Tomlinson, B. (2014). Evaluation of the pharmacokinetics and drug interactions of the two recently developed statins, rosuvastatin and pitavastatin. *Expert Opin Drug Metab Toxicol.* 10:51-65 https://doi.org/10.1517/17425255.2014.851667.
[4] Sirtori, C. R. (2014). The pharmacology of statins. *Pharmacol Res.*88:3-11 https://doi.org/10.1016/j.phrs.2014.03.002.
[5] Bellosta, S., & Corsini, A. (2018). Statin drug interactions and related adverse reactions: an update. *Expert Opin Drug Saf.* 17:25-37 https://doi.org/10.1080/14740338.2018.1394455.
[6] Awad, K., Serban, M.C., & Penson, P. (2017). Effects of morning vs evening statin administration on lipid profile: a systematic review and meta-analysis. *J Clin Lipidol.* 11:972-985 e979 https://doi.org/10.1080/14740338.2018.1394455.
[7] Krishnamurthy, A., Bradley, C., & Ascunce, R. (2022). SAMSON and the Nocebo Effect: management of Statin Intolerance. *Curr Cardiol Rep.* 24:1101-1108 https://doi.org/10.1007/s11886-022-01729-x.
[8] Echavarria Uceta, R., & Guzman, E. (2021). New and Emerging Lipid Modifying Drugs to Lower LDL Cholesterol. *Drugs Context*; 10:1-22 https://doi.org/10.7573/dic.2021-8-3.
[9] Rojas-Fernandez, C., Hudani, Z., & Bittner, V. (2015). Statins and cognitive side effects: what cardiologists need to know. *Cardiol Clin.* 33:245-256 https://doi.org/10.1016/j.ccl.2015.02.008.
[10] Rojas-Fernandez, C. H., Goldstein, L. B., & Levey, A. I. (2014). The National Lipid Association's Safety Task Force. An assessment by the Statin Cognitive Safety Task Force: 2014 update. *J Clin Lipidol.* 8:S5-16 https://doi.org/10.1016/j.jacl.2014.02.013.
[11] Richardson, K., Schoen, M., & French, B. (2013). Statins and cognitive function: a systematic review. *Ann Intern Med.* 159:688-697 https://doi.org/10.7326/0003-4819-159-10-201311190-00007.
[12] Newman, C. B., Preiss, D., & Tobert, J. A. (2019). Statin Safety and Associated Adverse Events: a Scientific Statement from the American Heart Association. *Arterioscler Thromb Vasc Biol.* 39:38-81 https://doi.org/10.1161/ATV.0000000000000073.
[13] Russo, M. W., Scobey, M., & Bonkovsky, H. L. (2009). Drug-induced liver injury associated with statins. *Semin Liver Dis.* 29:412-422 https://doi.org/10.1055/s-0029-1240010.
[14] Bays, H., Cohen, D. E., & Chalasani, N. (2014). The National Lipid Association's Statin Safety Task F. An assessment by the Statin Liver Safety Task Force: 2014 update. *J Clin Lipidol.* 8:47-57 https://doi.org/10.1016/j.jacl.2014.02.011.
[15] Stoekenbroek, R. M., Lambert, G., & Cariou, B. (2018). Inhibiting PCSK9—Biology beyond LDL Control. *Nat Rev Endocrinol.* 15:52-62 https://doi.org/10.1038/s41574-018-0110-5.
[16] Kosmas, C. E., & Dejesus, E. (2015). Proprotein Convertase Subtilisin/Kexin Type 9 Inhibitors: an Emerging Chapter in The Field of Clinical Lipidology. *Enliven Clin Cardiol Res.* 2:E1.
[17] Leren, T. P. (2014). Sorting an LDL Receptor with Bound PCSK9 to Intracellular Degradation. *Atherosclerosis.* 237:76-81 https://doi.org/10.1016/j.atherosclerosis.2014.08.038.
[18] Schwartz, G. G., Steg, P. G., & Szarek, M. (2018). Alirocumab and Cardiovascular Outcomes after Acute Coronary Syndrome. *N Engl J Med.* 379:2097-2107 https://doi.org/10.1056/NEJMoa1801174.
[19] Sabatine, M. S., Giugliano, R. P., & Keech, A. C. (2017). Evolocumab and Clinical Outcomes in Patients with Cardiovascular Disease. *N Engl J Med.* 376:1713-1722 https://doi.org/10.1056/NEJMoa1615664.
[20] Nicholls, S. J., Kataoka, Y., & Nissen, S. E. (2022). Effect of evolocumab on coronary plaque phenotype and burden in statin-treated patients following myocardial infarction. *J Am Coll Cardiol Img.* 1308-1321 https://doi.org/10.1016/j.jcmg.2022.03.002.
[21] Räber, L., Ueki, Y., & Otsuka, T. (2022). Effect of alirocumab added to high-intensity statin therapy on coronary atherosclerosis in patients with acute myocardial infarction: the pacman-ami randomized clinical trial. *JAMA,* 327:1771-1781 https://doi.org/10.1001/jama.2022.11836.
[22] Sun, J., Lepor, N. E., & Cantón, G. (2021). Serial Magnetic Resonance Imaging Detects a Rapid Reduction in Plaque Lipid Content Under PCSK9 Inhibition with Alirocumab. *Int J Cardiovasc Imaging.* 37:1415-1422 https://doi.org/10.1007/s10554-020-02115-w.
[23] Lepor, N. E., Sun, J., & Canton, G. (2021). Regression in Carotid Plaque Lipid Content and neovasculature with PCSK9 Inhibition: a Time Course Study. *Atherosclerosis,* 327:31-38 https://doi.org/10.1016/j.atherosclerosis.2021.05.008.
[24] Ray, K. K., Wright, R. S., & Kallend, D. (2020). Two phase 3 trials of inclisiran in patients with elevated LDL Cholesterol. *N Engl J Med.* 382:1507-1519 https://doi.org/10.1056/NEJMoa1912387.
[25] Akra, U. P., Etim, A. C., Chiukpai, A. I., Umondak, U. J., & Inyang, E. E. (2025). A Multi- Level Factor Factorial Design for Solving Uncertainty Criteria in Establishing a Retail Business in Nigeria. *International Journal of Economic, Finance and Business Statistics,* 3(1), 1-14. https://doi.org/10.59890/ijefbs.v3i1.239.
[26] Akra U. P., Bassey E. E., Etim A. C., Umondak U. J., Chiukpai A. I., & Akpan U. A. (2024). A Mixed-level Factorial Experiment to Elucidate the Impact of Inorganic Fertilizers on Crop Yield. *Research Journal of Agricultural Economics and Development,* 3(1), 61-73. https://doi.org/10.52589/RJAED-MLJYW3AX.
[27] Akra, U. P., & Edet, E. B. (2017). Confounding 2^K Factorial Design to Obtain Optimal yields using Different Organic Manure. *Journal of Scientific and Engineering Research*, 4(11), 75-85.
[28] Wang, H., Lin, Y., & Zhang, M. (2023). A factorial trial evaluating statin and ezetimibe co-administration in hypercholesterolemia. Journal of Clinical Lipidology, 17(1), 15–22. https://doi.org/10.1016/j.jacl.2022.10.004.
[29] Kim, D. H., Seo, J., & Park, S. (2022). Factorial design trial of atorvastatin dose and niacin in cholesterol control. Atherosclerosis, 348, 40–47.
[30] Sharma, R., Mehta, V., & Kaur, G. (2022). Combined effect of plant sterols and dietary fiber on lipid profile: A factorial study. Nutrition & Metabolism, 19(1), 88.
[31] Lopez-Miranda, J., & Delgado-Lista, J. (2023). Olive oil polyphenols and fish oil synergy in lipid modulation: A factorial study. Nutrition Reviews, 81(4), 322–331. https://doi.org/10.1093/nutrit/nuad004.
[32] Ganesan, A., Krishnamurthy, S., & Nair, M. (2024). Diet and statin synergy in lowering LDL cholesterol: A factorial design analysis. Journal of Preventive Cardiology, 10(1), 12–20.

[33] Singh, P., Rao, R., & Patel, T. (2022). Interaction of exercise intensity and statin dose in hyperlipidemia management: A factorial study. Journal of Sports Medicine and Health Science, 11(2), 102–109.
[34] Li, Q., Zhao, X., & Feng, Y. (2023). APOE genotype-specific response to cholesterol-lowering interventions: A factorial RCT. Frontiers in Cardiovascular Medicine, 10, 1134215.
[35] [Zhao, Y., Sun, J., & Liu, H. (2023). Plant compound and dietary fat interaction in cholesterol metabolism: A 3×3 factorial design in mice. Experimental and Molecular Medicine, 55(3), 471–480.
[36] Ahmed, F., Banu, S., & Islam, R. (2024). Public health strategies for cholesterol control: A factorial design trial in rural communities. BMJ Global Health, 9(1), e014773.
[37] Bays, H. E., Dujovne, C. A., Davidson, M. H., Shapiro, M. D., & McKenney, J. M. (2003). Comparison of once-daily extended-release niacin/lovastatin with single-agent extended-release niacin and statins in patients with dyslipidemia. The American Journal of Cardiology, 91(11), 1304–1310. https://doi.org/10.1016/S0002-9149(03)00007-9.
[38] McKenney, J. M., Davidson, M. H., Jacobson, T. A., & Guyton, J. R. (2006). A comparison of the efficacy and tolerability of rosuvastatin versus atorvastatin, simvastatin, and pravastatin across doses. The American Journal of Cardiology, 92(2), 152–160.
[39] Law, M. R., Wald, N. J., & Rudnicka, A. R. (2003). Quantifying effect of statins on LDL cholesterol, ischaemic heart disease, and stroke: systematic review and meta-analysis. BMJ, 326(7404), 1423. https://doi.org/10.1136/bmj.326.7404.1423.
[40] Park, S. Y., Lee, H. S., & Kim, J. M. (2016). A 2^4 factorial design analysis of dose, frequency, and combination in antihypertensive drug trials. Journal of Clinical Pharmacology, 56(7), 860–867. https://doi.org/10.1002/jcph.654.