

EFFECT OF POLOXAMER 407 ON SERUM VLDL, LDL AND HDL LEVELS OF RABBITS

Background: Poloxamer 407 is used in parenteral formulations as solubilizing and wetting agent for traditional, low-molecular-weight organic drug molecules and as stabilizing agent for proteins and polypeptide drugs. It has very high promising value in the medicine field, but has held responsible for changes in the lipid parameters. Thus, effect of poloxamer 407 on the rabbit's serum very low density lipoprotein (VLDL), low-density lipoprotein (LDL), and high-density lipoprotein (HDL) levels was studied after being injected intravenously.

Aims & Objective: To study the effect of poloxamer 407 on rabbit's serum VLDL, LDL, and HDL levels following intravenous injection.

Materials and Methods: Pretreatment, baseline readings were recorded. Rabbits were injected with 5.5, 27.5, and 137.5 mg/kg of poloxamer 407, and the effects on blood chemistry were assessed on the 2nd, 4th, and 7th day. The results of the study were expressed as mean \pm SEM, and data were analyzed using one-way analysis of variance test. Values with $P < 0.05$ were considered as significant.

Results: The highest dose of poloxamer 407 (137.5 mg/kg) significantly increased serum VLDL and decreased HDL level in rabbits, with the maximum increase observed on the 2nd day after injection. All the doses given did not alter the serum LDL level. The lower doses of poloxamer 407 did not alter serum VLDL, LDL, and HDL levels.

Conclusion: Our results showed that poloxamer 407 in higher doses significantly increased serum VLDL and decreased HDL levels but in lower doses did not show any adverse effect on the lipid biochemistry. But we recommend further studies to know the effect of this poloxamer on chronic use and through different route of administration.

Key Words: Poloxamer 407; Very Low Density Lipoprotein; Low-Density Lipoprotein; High-Density Lipoprotein; Rabbit

Harish Govind Naik¹, Ashwini Kolar²,
Dundesh Maled³, Chitra C Khanwelkar⁴,
Rohit Desai⁴, Sunil Gidamudi⁴

¹ Department of Pharmacology, Azeezia Medical College, Meeyanoor, Kollam, Kerala, India

² Department of Pathology, Azeezia Medical College, Meeyanoor, Kollam, Kerala, India

³ Department of Pharmacology, Institute of Medical Sciences & Research (IMSR), Mayani, Maharashtra, India

⁴ Department of Pharmacology, Krishna Institute of Medical Sciences Deemed University (KIMSUDU), Karad, Maharashtra, India

Correspondence to: Harish Govind Naik (harishhnaik@gmail.com)

Received Date: 26.05.2014

Accepted Date: 04.06.2014

DOI: 10.5455/njppp.2014.4.040620141

INTRODUCTION

Recently, there has been a gradual increase in mortality due to coronary heart diseases (CHDs). Factors such as hypercholesterolemia, cigarette smoking, diabetes mellitus, and sedentary lifestyle are key contributors to the development of hyperlipidemia and atherosclerotic cardiovascular disease. Apart from these factors, there are evidences that suggest certain chemicals can also cause hyperlipidemia and may eventually lead to CHDs. Poloxamer 407 is one such example.^[1]

Poloxamer 407 is a hydrophilic nonionic surfactant that belongs to a class of copolymers called poloxamers. It is a triblock copolymer that displays concentration-dependent reverse thermal gelation, a characteristic that makes it potentially useful for the development of the sustained-release injectable drugs.^[2] This property is of prime interest in optimizing drug formulation.

Poloxamer 407 is now widely preferred as solubilizing and wetting agent in parenteral formulations for low-molecular-weight organic drug molecules. It is also

preferred as stabilizing agent for proteins and polypeptide drugs.^[3] Solubilization of poorly water soluble drugs is enhanced by poloxamer 407 formulation. It is also helpful in prolonging the release profile of many galenic applications.^[4]

Poloxamer 407 shows thermoreversible properties and undergoes reverse thermal gelation; it can be injected as a solution but then forms a gel matrix at body temperature.^[4]

Researches to develop a sustained-release formulation of recombinant human growth hormone (rhGH) are conducted and have shown that the controlled-release formulation continued to release rhGH *in vitro* for 60–72 hours and *in vivo* for a week following intramuscular and subcutaneous injections.^[5]

Intravenous injection of acyclovir sodium produces phlebitis at the injection site, which is one of the most frequent adverse reactions reported. Acyclovir nanoparticle made from poloxamer 407 has reduced the incidences of thrombophlebitis caused by parenteral

acyclovir. The presence of poloxamer 407 in the nanoparticles is responsible for enhancing the release of acyclovir.^[6]

Apart from the above-mentioned useful properties, poloxamer 407 has immunomodulation and cytotoxicity-promoting properties; this has significantly increased the pharmacological interest of poloxamer 407. Therefore, human trials are in progress to specify these potential applications.

Poloxamer 407 has very high promising value in the medicine field, but has held responsible for changes in the lipid parameters.^[4] This may hinder the development of various parental formulations using poloxamer 407. Hence this study was proposed to evaluate the effect of poloxamer 407 on rabbit's serum lipoprotein levels after single intravenous injection. In case of any alteration in the lipoprotein levels, further studies involving multiple doses of poloxamer 407 can be planned.

MATERIALS AND METHODS

All experimental protocols involving animals were followed in accordance with the guidelines from the Committee for the Purpose of Control and Supervision of Experiments on Animals. The study designs were evaluated and were approved by the institutional animal ethical committee of Krishna Institute of Medical Sciences Deemed University (KIMSUDU), Karad, Maharashtra.

Laboratory-bred rabbits of either sex (2000 ± 200 g) were maintained under standard laboratory conditions (temperature $20 \pm 2^\circ$ C, 12 h light/dark cycle). They were provided water and food *ad libitum*. The experiments were conducted under aseptic conditions in the laboratory of the Department of Pharmacology, KIMSUDU.

Chemical: Poloxamer 407 was a gift from BASF India Limited.

Fifteen rabbits of either sex were divided into three groups, having five rabbits in each group.

Pretreatment, baseline serum very low density lipoprotein (VLDL), low-density lipoprotein (LDL), and high-density lipoprotein (HDL) levels of rabbits were estimated by collecting blood samples from the marginal ear vein.

Poloxamer 407 was given as single intravenous dose to

rabbits as follows:

- Group I ($n = 5$): 5.5 mg/kg body weight
- Group II ($n = 5$): 27.5 mg/kg body weight
- Group III ($n = 5$): 137.5 mg/kg body weight

The rabbits were held using the rabbit holder with the help of the animal house staff of the institute. The dose was administered as a bolus injection to the respective groups through the marginal vein of the ear.

Collection of Blood: Samples of blood (1 ml) were withdrawn from the marginal vein of the other ear at different time intervals on the 2nd, 4th, and 7th day following poloxamer 407 intravenous injection.

Biochemical Analysis: Serum VLDL, LDL, and HDL levels were estimated in the biochemistry laboratory of the institute.

Statistical Analysis: The results of the study were expressed as mean \pm SEM. Data were analyzed by using one-way analysis of variance test. Values with $P < 0.005$ were considered as significant.

RESULTS

Animals were observed for 30 minutes after the intravenous injection of poloxamer 407. In all the administered doses of poloxamer 407, no death was observed.

Table-1: Effect of Poloxamer 407 on Rabbit's Serum VLDL, LDL, and HDL Levels

Group	Dose of Poloxamer 407 (mg/kg)	Level of Lipoproteins			
		Before Rx	After Rx		
			Day 2	Day 4	Day 7
VLDL					
I	5.5	28.2 \pm 7.59	30.2 \pm 7.65	28.6 \pm 8.76	35.4 \pm 11.45
II	27.5	23.6 \pm 6.74	24.6 \pm 8.23	19.6 \pm 5.87	22.8 \pm 6.14
III	137.5	36.4 \pm 5.06	63.8 \pm 12.96	35.2 \pm 5.38	30.8 \pm 5.02
LDL					
I	5.5	21.3 \pm 7.4	21 \pm 7.45	18.8 \pm 5.32	18.4 \pm 5.98
II	27.5	12.4 \pm 3.82	13.4 \pm 3.5	11.8 \pm 2.31	8.72 \pm 1.89
III	137.5	19.4 \pm 6.79	16.8 \pm 5.53	17 \pm 5.25	19.4 \pm 5.35
HDL					
I	5.5	11.9 \pm 3.1	13 \pm 3.05	12 \pm 2.34	12.6 \pm 2.73
II	27.5	9.6 \pm 2.8	7.8 \pm 0.98	7.4 \pm 1.43	5.78 \pm 1.12
III	137.5	6.6 \pm 0.74	2.08 \pm 0.72	4.04 \pm 0.8	5.66 \pm 1.16

Values are expressed as mg/dl, mean \pm SEM. * $P < 0.05$ (ANOVA) statistically significant.

Administration of poloxamer 407 intravenously led to the decrease of serum HDL level and increase of serum VLDL level in Group III, which was treated with a dose of 137.5 mg/kg body weight.

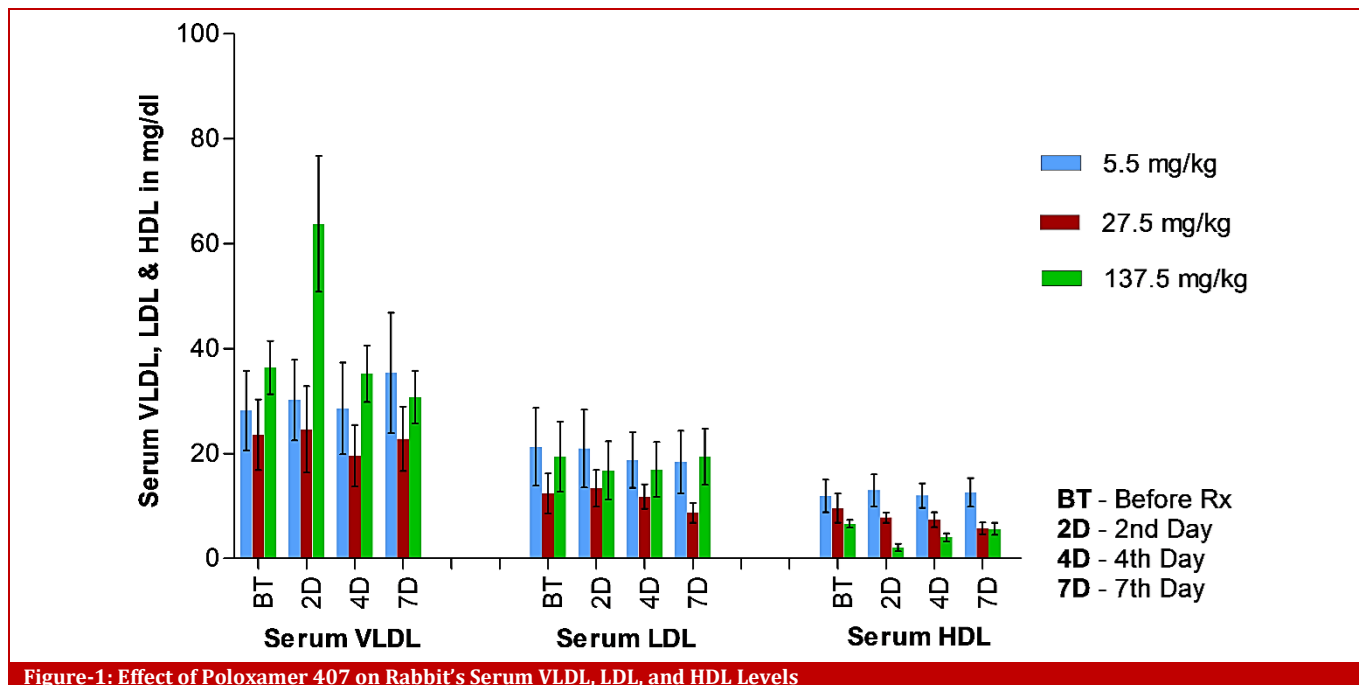


Figure-1: Effect of Poloxamer 407 on Rabbit's Serum VLDL, LDL, and HDL Levels

Serum HDL level of Group III showed variation among the means at various time intervals, which was found to be significantly higher than expected ($P < 0.05$). It is observed that on the 2nd day mean serum HDL level decreased whereas the mean serum VLDL level increased significantly. The levels of VLDL and HDL cholesterol did not increase significantly in other groups (i.e., Groups I and II). The level of LDL cholesterol did not change significantly in all the groups.

DISCUSSION

Statistically significant increase in serum VLDL level and decrease in serum HDL level in Group III (poloxamer 407 in a dose of 137.5 mg/kg) were observed. The concept that CHD can be prevented has increasingly become a driving force in cardiovascular medicine. Widespread acceptance of the benefits of prevention came first in the area of secondary prevention. Many cardiologists consider secondary prevention to be treatment of coronary artery disease.^[7] The lipid-atherogenesis connection has undergone a major change from an initial focus on serum total cholesterol, later on the other lipids and then on the lipoproteins that transport them, and now on the distribution of cholesterol in the LDLs, HDLs, and VLDLs.^[8] Elevated serum LDL and VLDL cholesterol, and low HDL cholesterol are the major risk factors for the CHD.^[7] In a large Chinese cohort, elevated VLDL cholesterol was found to be significantly associated with elevated CHD risk, similar to that observed for LDL cholesterol.^[9] Studies suggest that VLDL receptor is an important contributor to the development of Alzheimer's

disease.^[10] HDL cholesterol is considered "good" cholesterol because it helps remove LDL cholesterol from the arteries. It acts as a scavenger, carrying LDL cholesterol away from the arteries and back to the liver, where it is broken down and passed from the body. One-fourth to one-third of blood cholesterol is carried by HDL. A healthy level of HDL cholesterol may also protect against heart attack and stroke, whereas low levels of HDL cholesterol have been shown to increase the risk of heart disease.^[11]

The alterations in the levels of serum VLDL and HDL cholesterol were found to be statistically significant on the 2nd day in Group III but were not statistically significant on the other days (i.e., 4th and 7th day). These alterations were recorded following single intravenous injection of poloxamer 407. It is desirable to know the alteration in the serum VLDL, LDL, and HDL cholesterol level that might occur following chronic exposure to poloxamer 407. We also recommend that further studies should be carried out to know the effect of poloxamer 407 following different routes of administration. A limitation of this study is that the changes in the lipid biochemistry are observed after only a single exposure to poloxamer 407. No statistically significant alteration in the LDL cholesterol level is observed in all three groups. The lower doses of poloxamer 407 did not alter the levels of serum VLDL, LDL, and HDL cholesterol, thus have not shown any adverse effect on the lipid profile.

CONCLUSION

Our results have shown that higher doses of poloxamer 407 significantly increased serum VLDL level and decreased serum HDL level but low doses had no adverse effect on the lipid biochemistry. We recommend further studies to know the effect of this poloxamer on chronic administration and also when administered through different routes.

ACKNOWLEDGEMENT

We acknowledge the support of the Krishna Teaching Hospital, Karad, Maharashtra.

REFERENCES

1. Wasan KM, Subramanian R, Kwong M, Goldberg IJ, Wright T, Johnston TP. Poloxamer 407-mediated alterations in the activities of enzymes regulating lipid metabolism in rats. *J Pharm Pharm Sci* 2003;6(2):189-97.
2. Blonder JM, Baird L, Fulfs JC, Rosenthal GJ. Dose-dependent hyperlipidemia in rabbits following administration of poloxamer 407 gel. *Life Sci* 1999;65(21):PL261-6.
3. Wout ZG, Pec EA, Maggiore JA, Williams RH, Palicharla P, Johnston TP. Poloxamer 407-mediated changes in plasma cholesterol and triglycerides following intraperitoneal injection to rats. *J Parenter Sci Technol* 1992;46(6):192-200.
4. Dumortier G, Grossiord JL, Agnely F, Chaumeil JC. A review of

- poloxamer 407 pharmaceutical and pharmacological characteristics. *Pharm Res* 2006;23(12):2709-28.
5. Katakam M, Ravis WR, Banga AK. Controlled release of human growth hormone in rats following parenteral administration of poloxamer gels. *J Control Release* 1997;49(1):21-6.
 6. Kamel AO, Awad GAS, Geneidi AS, Mortada ND. Preparation of intravenous stealthy acyclovir nanoparticles with increased mean residence time. *AAPS PharmSciTech* 2009;10(4):1427-36.
 7. Grundy SM. Primary prevention of coronary heart disease integrating risk assessment with intervention. *Circulation* 1999;100(9):988-98.
 8. Kannel WB. High-density lipoproteins: epidemiologic profile and risks of coronary artery disease. *Am J Cardiol* 1983;52(4):9B-12B.
 9. Ren J, Grundy SM, Liu J, Wang W, Wang M, Sun J, et al. Long-term coronary heart disease risk associated with very-low-density lipoprotein cholesterol in Chinese: the results of a 15-year Chinese Multi-Provincial Cohort Study (CMCS). *Atherosclerosis* 2010;211(1):327-32.
 10. Okuizumi K, Onodera O, Namba Y, Ikeda K, Yamamoto T, Seki K, et al. Genetic association of the very low density lipoprotein (VLDL) receptor gene with sporadic Alzheimer's disease. *Nat Genet* 1995;11(2):207-9.
 11. American Heart Association. Good vs Bad Cholesterol. Dallas, TX, American Heart Association, 2014. Available from URL: http://www.heart.org/HEARTORG/Conditions/Cholesterol/AboutCholesterol/Good-vs-Bad-Cholesterol_UCM_305561_Article.jsp [Accessed May 20, 2014].

Cite this article as: Naik HG, Kolar A, Maled D, Khanwelkar CC, Desai R, Gidamudi S. Effect of Poloxamer 407 on serum VLDL, LDL and HDL levels of rabbits. *Natl J Physiol Pharm Pharmacol* 2014; 4:221-224.

Source of Support: Nil

Conflict of interest: None declared