Original Article

Effect of Cinnamomum cassia Bark Extract, Pyridoxine and Pitavastatin on Diet-Induced Murine Hypertriglyceridemia and Obesity

Maryam Mansoor, Saadia Shahzad Alam, Iram Imran, Talha Laique and Kamran Dawood Ahmad

Objective: To compare effects of aqueous Cinnamomum cassia bark extract (ACCE), pyridoxine (PYR) and pitavastatin (PIT) and dietary prevention on TG and body weight of rats.

Methods: Sixty albino male rats: sorted into 10 groups (Group 1 - control). Group 2 (dietary preventive) and Groups 3-10 (therapeutic) were induced using HFD for 30 days (HFD continued throughout). Group 2 was later reverted to normal diet. They were treated post-induction for 30 days, once daily: Group 3 (PIT 0.3mg/kg); Group 4 (PYR 18mg/kg), Group 5 (ACCE 200mg/kg), Group 6 (PIT 0.3mg/kg + PYR 18mg/kg), Group 7 (PYR 18mg/kg + ACCE 200mg/kg), Group 8 (PIT 0.3mg/kg + ACCE 200mg/kg), Group 9 (PIT 0.3mg/kg + PYR 18mg/kg + ACCE 200mg/kg) and Group 10 (PIT 0.15mg/kg + PYR 9mg/kg + ACCE 100mg/kg). TG and weight was checked (Day 0, Day 30, Day 60) and data analyzed using SPSS 20.0 ($P \leq 0.05$).

Results: Rats showed significant reduction in serum TG after treatment. Maximum effect was seen in Group 7 (PYR 18mg/kg + ACCE 200mg/kg) where TG levels decreased to 94.667 ± 20.077 mg/dL. Dietary alteration in Group 2 resulted in only marginal improvement. Weight reduction of 23 gm was seen in Group 4 (ACCE 200mg/kg).

Conclusions: Cinnamon, pyridoxine and pitavastatin showed remarkable TG-lowering activity. PYR and ACCE exhibited additive effect (39%), endorsing the need for "integrated" approach. Cinnamon can treat obesity in humans.

Keywords: hypertriglyceridemia, cinnamon, pyridoxine, pitavastatin, high-fat diet.

Introduction

Hypertriglyceridemia (HTG), known as "hyperlipemia", a common disorder all over the world, is a condition of raised serum triglycerides. In experimental and epidemiologic studies, hyperlipemia is a strong risk factor for cardiovascular disorders especially ischemic heart diseases (IHDs) especially coronary artery diseases (CAD).[1] It can be classified into primary and secondary hypertriglyceridemia. Primary hypertriglyceridemia is mostly genetic.[2] Secondary hypertriglyceridemia is however, acquired: caused or exacerbated by obesity, diabetes mellitus, endocrine disorders, diet and lifestyle, all are prevalent in more modernized societies.[1] Based on NCEP (National Cholesterol Education Program) ATIII Guidelines, HTG is defined as a fasting serum triglyceride level greater than 200 mg/dL.[3] Obesity has been found to be linked to HTG. Both are often found together in an ominous picture of "metabolic syndrome", together with other disorders. Obesity is a complex multi-factorial (genetics, environment) chronic metabolic disorder. Its prevalence is increasing day by day, making it the second leading cause of morbidity and mortality globally. It has been estimated to cause approximately 2.6 million deaths worldwide.[4] Mild HTG can be controlled by lifestyle modification: dietary counselling and increasing physical activity to achieve weight reduction in overweight/obese patients, linking it to obesity. Severe HTG has an underlying etiology of commonly, an amalgam of both factors: genetic and secondary. Family history of dyslipidemia and cardiovascular diseases for future risk assessment,[5,6] history of alcoholism and use of notorious medications should be probed. Physical findings pan across multi-organ systems and hospital admission for aggressive medical treatment is necessary (abdominal pain/pancreatitis).[6]

Treatment of severe HTG necessitates the use of pharmacologic agents. Intravenous insulin and rarely, plasma exchange may be needed if oral therapy fails. Patients should be assessed for other CV risk factors e.g. central obesity, hypertension, glucose metabolism abnormalities and liver disorders. Niacin, fibrates and omega-3 fatty acids with or without statins (reductase inhibitors) are routinely used. Statins can thus be useful in treating mild-moderate HTG alone, especially when indicated to decrease cardiovascular risk.[5] The model of HTG used in this study comes under mild HTG. Hence they are prescribed worldwide for risk reduction of CVDs. Thus we have

used pitavastatin, which is a strong inhibitor of HMG-CoA reductase and lower serum lipids especially TC, LDL-C and TGs.[7] Pharmacological treatment has its advantages and disadvantages, both. So, emphasis has been made on the utilization of phyto-alternatives for treatment of various ailments.[8] Plants and plant products are becoming first choice of masses of the developing world owing to their reputation of being effective, safe and affordable.[10] Their use has also been penned down in various pharmacopoeias.[9] Experimentation with plants have shown promising results in rats, e.g. Brassica compestris (mustard), Trigonella feonum-graecum (fenugreek) and Cinnamomum cassia (Cinnamon), etc. lower serum TGs.[11] Cinnamomum cassia especially bark extracts, have traditionally been used in treating lipid-related disorders due to its anti-hyperlipidemic activity.[12]

Researchers have claimed that it reduces lipid levels notably total cholesterol, TGs and LDL substantially in experimental animals as well as humans; treats diabetes and obesity, and stabilizes blood pressure.[12,13] Several micronutrients have also been found useful in the treatment of dyslipidemia especially niacin, cobalamin,[14,15] lysine, pyridoxine and more.[11] Pyridoxine (Vitamin B_6)is present in the body in interconvertible "alcohol", "aldehyde" and "amine" forms.[16] It shows pronounced effect in decreasing TGs by affecting mobilization of unsaturated FAs from TGs to phospholipids.[17]

There is a need to find out about which approach to the treatment of hypertriglyceridemia (HTG) and obesity is more beneficial: pharmacotherapy, phytotherapy or micronutrient approach. Also, more studies are needed be conducted to find out whether they could or couldn't be used in combination and that which combination would be the most efficacious. Keeping this in view, the study was designed to investigate the most efficacious options out of pitavastatin, cinnamon or pyridoxine in treating HTG and obesity in rats.

Methods

Quasi-experimental animal trial & simple random sampling; n=6. Pitavastatin (2mg) tablets, Pyridoxine (25mg) tablets were purchased from Clinix Pharmacy, Lahore. Cinnamon bark was purchased from Hamdard Dawakhana, Lahore.1kg Cinnamomum cassia bark was cleaned, dried in a shady place and extracted with 4L of distilled water at 90°C for 16 hours, twice. Extract was filtered and then freeze-dried for storage and preservation at room temperature till use; ready to be diluted in normal saline (0.9%) and orally administered at 200mg/kg. Dry yield was 8% (w/w).[18] The extract was prepared at Pharmacology Laboratory, UHS, Lahore. 60 healthy young male albino rats (6 weeks of age), weighing 150-170 grams, were purchased from UHS, Lahore and were kept in polypropylene cages at UHS under standard lighting & housing conditions; were fed laboratory prepared diet and tap water, ad libitum. Controls received normal diet. In experimental groups, High-Fat Diet (HFD) was used for induction of hypertriglyceridemia and obesity for 30 days. HFD **(Table-1)** was purchased from University of Veterinary and Animal Sciences, Lahore. Group 2 received HFD for only 30 days and was later on, reverted to normal laboratory diet from Day 30-Day 60 (end of the study period) and left untreated to study preventive role of dietary modification. The rest of the groups 3-10 were continued on HFD throughout along with treatment.[19] In this study, 10 groups of 6 animals each were used **(Table -1)**. Body weight and TGs of the rats were checked at Day 0, Day 30 and Day 60; data analyzed using SPSS 20.0 ($P \leq 0.05$). Weight was estimated using electronic weighing balance. Blood samples for TGs were collected via cardiac puncture technique into serum

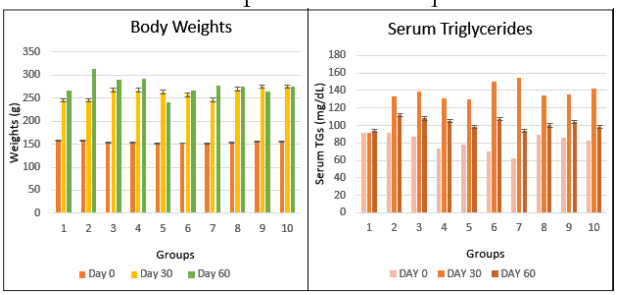

Fig-1: Comparison of weights& triglycerides at day 0, 30 and 60.

Table-1: Groups Distribution and constituents of HFD.

Group	Distribution
Group 1	Control
Group 2	(Preventive Model - HFD + Normal Diet)
Group 3	(HFD + 0.3mg/kg Pitavastatin)
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Constituents of HFD: Casein: 120g; Corn starch: 549.6g: Soybean oil: 250g; Cholesterol: 10; Choline: 0.4g; Salt mixture: 50g; Vitamin mixture: 10g; Cellulose: 10g; Total calories (Kcal)-5018.4/1000g of diet

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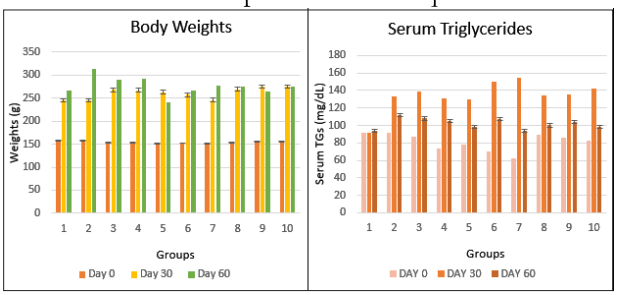

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lowering effect seen in Group 2 (preventive) validating the inadequacy of preventive strategy alone in handling hyperlipemia. Groups 6(PIT 0.3mg/kg+PYR 18mg/kg), Group 7(PYR 18mg/kg+ACCE 200mg/kg) and Group10(PIT 0.15mg/kg+PYR 9mg/kg+ACCE 100mg/kg) revealed the most favorable hypotriglyceridemic effect of 28.6% (107mg/dL from 150mg/dL), 39% (94mg/dL from 155mg/dL) and 31% (98mg/dL from 142mg/dL),respectively. As pyridoxine was found constant in every group, it can be deduced that pyridoxine showed a "synergistic" effect on serum TGs in each combination with statin and cinnamon, even when their doses were halved (Group 10). No previous study exists to support their synergism. The potent TG-lowering effect of these agents, individually, was also supported by previous studies especially pyridoxine. Pyridoxine is known to affect methylation of phospholipids, desaturation and elongation of FAs and mobilization of unsaturated FAs from TGs to phospholipids.[17] Cinnamon acts like a dual activator of PPAR-ɣ and PPAR-α,[25] which regulate metabolism of blood glucose and lipids, respectively. As a vital part of its effect on metabolism of carbohydrates, it enhances insulin sensitivity. And one of the important actions of insulin is to promote deposition of lipids into the stores (as TGs) and increase their clearance from the blood. That is why TG levels fall rapidly when cinnamon is administered.[26]

So, it can be summarized that pyridoxine and cinnamon, both have beneficial anti-obesity and TG-lowering effects comparable to any standard pharmacological treatment (pitavastatin in this case). As recently, people's trust on plant products and micronutrients seems to be regained, other beneficial outcomes of the use of these agents need to be explored and assayed with routinely used pharmacological agents

Conclusion

Cinnamon, pyridoxine and pitavastatin remarkably decreased serum triglycerides, individually as well as in combinations, which signify their additive effects. Dietary prevention in the absence of treatment also plays a minor role. Cinnamon's beneficial role in treating obesity has also been evident as it alone reduced body weight of the rats, despite their consumption of high-fat diet throughout the study period. Hence, micronutrients and phyto-alternatives can also be utilized for the treatment of hypertriglyceridemia and obesity in humans along with pharmacological agents

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