# Comparative evaluation between chitosan and atorvastatin on serum lipid profile changes in hyperlipidemic cats

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(Received 20 Jul 2015; revised version 19 Dec 2015; accepted 28 Dec 2015)

## Summary

The purpose of the present survey was to determine the effects of the chitosan and atorvastatin on serum lipid profile changes and the influence of time on treatment process in cats. For the management of cholesterol induced hyperlipidemia, twenty-one healthy cats were randomly divided into three equal groups. Group A (control) included seven cats that were fed with cholesterol powder (4 g/kg for 10 days). Group B was similar to group A, but in addition, atorvastatin (5 mg/kg) was administered for 45 days after induced hyperlipidemia. Group C was similar to group B, but chitosan (3 g/cat) was administered instead of atorvastatin. Blood samples were collected four times on days 0, 10, 40 and 55 after challenge. Serum total cholesterol, triglycerides, HDL-C and LDL-C levels were measured using standard commercial kits. Atorvastatin ($P<0.001$) and chitosan ($P<0.01$) showed more hypolipidemic activity in lowering triglycerides compared with group A. In a comparison between two drugs and their effects on triglyceride, atorvastatin showed a significant difference with chitosan ($P<0.01$). Atorvastatin ($P<0.01$) and chitosan ($P<0.05$) showed more activity in lowering cholesterol than the control group. The treated groups (B and C) had good results in lowering LDL-C, compared with group A, on day 45 ($P<0.001$). A significant difference was seen only between groups A and B and on day 45 in increase of HDL-C ($P<0.01$). In conclusion, it was shown that although both drugs had hypolipidemic activity in cats, atorvastatin was more effective than chitosan. Further experimentation will be needed to elucidate the possible biochemical mechanism of the drugs.

**Key words:** Atorvastatin, Cat, Chitosan, Hyperlipidemia, Lipid profiles

## Introduction

The term hyperlipidemia is defined as an elevation of one or more of the serum lipids, including cholesterol, cholesterol esters, triglycerides and phospholipids (Johnson, 2005). Lipemia is apparent when serum triglyceride concentrations exceed 2.26 mmol/L (200 mg/dl) in dogs and cats (Bauer, 2004). As serum triglyceride concentration increases, serum becomes turbid and then lactescent (Johnson, 2005). Lipoproteins can be divided based on their hydrated density into four major classes: chylomicrons, very low-density lipoproteins (VLDL), low-density lipoproteins (LDL), and high-density lipoproteins (HDL) (Maldonado *et al*., 2001). The most characterized primary lipid disorder in cats is inherited fasting hyperchylomicronemia, an autosomal recessive disorder, resulting from decreased lipoprotein lipase activity (Ross *et al*., 2006). It has been shown that 25% of randomly selected Burmese cats in Australia exhibit marked post-prandial hyper-triglyceridemia after an oral fat tolerance challenge (Kluger *et al*., 2010).

Hyperlipidemia can be either primary or secondary to other diseases. Primary lipid disorders are not commonly observed in cats. Secondary hyperlipidemia has been reported in some diseases such as diabetes mellitus, idiopathic hyperlipidemia, pancreatitis, nephrotic syndrome and cholesterol ester storage disease. Another manifestation of lipid disorders in cats is lipid aqueous, a sporadic condition whereby lipid accumulates in the aqueous humor of the eye. This condition has been described in Burmese and Tonkinese cats in Australia and the United Kingdom (Kluger *et al*., 2009; Xenoulis and Steiner, 2010).

The first step in the treatment of hyperlipidemia is to determine whether the animal has a primary or a secondary lipid disorder. Cats with primary hyperlipidemia should be offered a low-fat diet throughout their lives. Diets that contain less than 20 g of fat per 1000 kcal are recommended (Elliott, 2005). Medical management is achieved by administration of low-fat diets with lipid-lowering agents such as atorvastatin, omega-3 fatty acids, gemfibrozil, garlic and niacin (Saravanan and Prakash, 2004; Kojuri *et al*., 2007; Allen Last *et al*., 2011). They are administered to treat hyperlipidemia, atherosclerosis or cardiovascular complications like coronary heart disease. Among the available 3-hydroxy-3-methyl-glutaryl-coenzyme A (HMG-CoA) reductase inhibitors, atorvastatin is one of the major lipid drugs to reduce the elevated lipid profiles in hyperlipidemic conditions (Tuccori *et al*., 2014). Although statins consistently reduce plasma LDL-C and total cholesterol levels by inhibiting the synthesis of cholesterol, emerging evidence suggests that statins have additional benefits in lowering the risk of a blood clot (Verd *et al*., 1999; Briand *et al*., 2006b).

Chitosan is a natural compound which professes to bind fats in the digestive tract. It is very safe, but efficacy is unproven in many animals. Chitosan should not be administered at the same time as fat soluble vitamins or therapeutic lipids such as fish oil. The mechanism of drug action may be due to the antioxidant and antilipid peroxidation (Xing *et al*., 2005; Choi *et al*., 2012). In recent years, international studies have revealed that chitosan is an ideal bioactive substance and an effective neuroprotection, anti-cancer, antibacterial, anti-inflammatory, hypoglycemic, antioxidant, and liver protection agent. In addition, chitosan is used in the field of food and nutrition to reduce lipid levels, serving as an effective lipid-lowering dietary supplement (Ju *et al*., 2010; Younes and Rinaudo, 2015). Studies evaluating the efficacy of chitosan in cats with hyperlipidemia are lacking and clinical experience is limited, so the purpose of the present survey was to evaluate and compare the effects of the chitosan and atorvastatin on serum lipid profile changes and the influence of time on treatment process in cats.

## Materials and Methods

Twenty-one healthy male cats, ages 1-2 years old, domestic short hair (DSH) breed and weighing 2.95-3.75 kg were selected and allowed to acclimatize to the environment for 14 days in separate cages. They were fed with a standard diet (chicken) and water was *ad libitum*. Determination of the age was accomplished based on dental formulary. For the management of cholesterol induced hyperlipidemia, the studied cats were randomly divided into three equal groups.

Group A (control): Included seven cats that were fed with cholesterol powder with dosage 4 g/kg for 10 days to induce hyperlipidemia (Batch No.: C-102; Chemsworth, India) (Sakamoto *et al*., 1991).

Group B: Similar to group A, but in addition, atorvastatin (Poursina Co., Tab 40 mg) was administered with dosage 5 mg/kg and for 45 days after induced hyperlipidemia.

Group C: Similar to group B, but chitosan (Sigma-Aldrich, Germany) was administered with dosage 3 g/cat instead of atorvastatin.

The selected dose for chitosan (medium molecular weight) was based on the used doses in human studies. Blood samples were collected four times on days 0, 10, 40 and 55 after challenge, to measure serum lipid profiles in the studied cats. The samples were poured into non-heparinized tubes and serum was obtained using centrifuge at 3000 rpm for 5 min at room temperature. Serum total cholesterol, triglyceride and LDL-C levels were determined using respective diagnostic commercial kits (Pars Azmoon and photometric method). HDL-C was measured using Pishtaz-Teb kit and direct method also.

### Statistical analysis

All data are presented as the means±standard deviation (SD). Statistical comparisons among therapeutic different groups (A, B and C) were carried out using one-way ANOVA test using SPSS for Windows, version 8.0 followed by Bonferroni's test. Differences implied statistical significance below $P<0.05$.

## Results

Atorvastatin ($P<0.001$) and chitosan ($P<0.01$) showed more hypolipidemic activity in lowering triglyceride compared with group A. In comparison between two drugs and their effects on triglyceride, atorvastatin showed a significant difference than chitosan ($P<0.01$). After 45 days of treatment with atorvastatin and chitosan drugs, triglyceride level decreased from 293.80 ± 16.33 and 270.40 ± 21.18 to 108.60 ± 6.88 and 170.40 ± 2.46 in groups B and C, respectively. Concerned with the effects of drugs on cholesterol level, atorvastatin ($P<0.01$) and chitosan ($P<0.05$) showed more activity in lowering cholesterol than the control group. In comparison between two drugs and their effects on cholesterol, a significant difference was seen between atorvastatin and chitosan on day 30 ($P<0.05$), but the difference was not significant on day 45. Total cholesterol levels reduced from 294.20 ± 7.84 and 287.60 ± 13.81 to 107.60 ± 5.15 and 185.20 ± 5.77 after 45 days of treatment, in groups B and C, respectively. The treated groups (B and C) had good results in lowering LDL, compared with group A, on day 45 ($P<0.001$). In comparison between two drugs and their effects in increase of HDL, a significant difference was seen between groups A and B on day 45 ($P<0.01$). In the present survey, cats fed with cholesterol powder, developed hyperlipidemia (increase of triglyceride or cholesterol) in 90.48% (19 out of 21 cases) of the treated cats. Two cats were resistant to the effects of cholesterol in induced hyperlipidemia, so two others replaced them. Normal levels were considered between 25-160 and 75-220 mg/dl for triglyceride and cholesterol, respectively (Elliott, 2005). The enhancement effects of chitosan and atorvastatin are clearly demonstrated in Tables 1, 2 and 3 in lowering the serum lipid profile parameters on days 0, 10, 40 and 55 after induced hyperlipidemia.

**Table 1:** Changes of serum lipid profile parameters (mg/dl) on days 0, 10, 40 and 55 after induced hyperlipidemia in cats of group A (n=7)

| Lipid profile parameters | Day 0 | Day 10 | Day 40 | Day 55 |
|---|---|---|---|---|
| Total cholesterol | $133.80 \pm 8.74^{a}$ | $289.20 \pm 7.54^{b}$ | $251.20 \pm 5.94^{b}$ | $224.80 \pm 2.85^{b}$ |
| Triglyceride | $101.80 \pm 5.99^{a}$ | $297.60 \pm 8.41^{b}$ | $260.20 \pm 15.00^{b}$ | $248.80 \pm 17.90^{b}$ |
| HDL-C | $59.00 \pm 6.44^{a}$ | $123.20 \pm 12.18^{b}$ | $102.00 \pm 6.77^{b}$ | $102.60 \pm 3.89^{b}$ |
| LDL-C | $37.40 \pm 3.59^{a}$ | $140.20 \pm 15.63^{b}$ | $92.20 \pm 5.58^{b}$ | $93.20 \pm 4.03^{b}$ |

Significant differences are presented by lowercase letters in each column ($P<0.05$)

**Table 2:** Effect of atorvastatin on serum lipid profile parameters (mg/dl) on days 0, 10, 40 and 55 after induced hyperlipidemia in cats of group B (n=7)

| Lipid profile parameters | Day 0 | Day 10 | Day 40 | Day 55 |
|---|---|---|---|---|
| Total cholesterol | 123.00 ± 2.98[a] | 294.20 ± 7.84[b] | 165.80 ± 11.24[c] | 107.60 ± 5.15[d] |
| Triglyceride | 119.60 ± 8.83[a] | 293.80 ± 16.33[b] | 155.40 ± 3.19[c] | 108.60 ± 6.88[d] |
| HDL-C | 78.80 ± 5.21[a] | 121.20 ± 3.14[b] | 99.60 ± 2.54[b] | 135.60 ± 10.99[c] |
| LDL-C | 40.00 ± 3.71[a] | 171.40 ± 8.51[b] | 56.00 ± 3.41[c] | 41.00 ± 2.88[d] |

Significant differences are presented by lowercase letters in each column (P<0.05)

**Table 3:** Effects of chitosan on serum lipid profile parameters (mg/dl) on days 0, 10, 40 and 55 after induced hyperlipidemia in cats of group C (n=7)

| Lipid profile parameters | Day 0 | Day 10 | Day 40 | Day 55 |
|---|---|---|---|---|
| Total cholesterol | 145.40 ± 10.63[a] | 287.60 ± 13.81[b] | 216.60 ± 5.28[c] | 185.20 ± 5.77[d] |
| Triglyceride | 139.20 ± 4.60[a] | 270.40 ± 21.18[b] | 204.00 ± 9.34[c] | 170.40 ± 2.46[d] |
| HDL-C | 85.80 ± 6.17[a] | 124.80 ± 4.44[b] | 114.00 ± 4.69[c] | 121.40 ± 6.01[c] |
| LDL-C | 46.80 ± 3.84[a] | 176.40 ± 10.40[b] | 71.80 ± 4.37[c] | 64.80 ± 3.34[d] |

Significant differences are presented by lowercase letters in each column (P<0.05)

## Discussion

In the present survey, the effects of chitosan and atorvastatin were evaluated on serum lipid profile changes in hyperlipidemic cats. The results showed that atorvastatin and chitosan both had hypolipidemic activity in cats, but atorvastatin was more effective than chitosan. The onset of hypolipidemic effects were evident as early as 30 days and became more progressive and greater after 45 days of treatment, in both groups B and C. In comparison between two drugs and their effects on triglyceride level, atorvastatin showed a significant difference than chitosan. Also, atorvastatin and chitosan showed more activity in lowering total cholesterol than the control group.

Atorvastatin has some side effects in human such as constipation, diarrhea, nausea, muscle pain, fever, dark urine, increased thirst, drowsiness, loss of appetite and yellowing of the skin or eyes (Ford, 1996). In dogs, the drug may cause multifocal hemorrhage in the sub-mucosa of the gallbladder and increase of liver enzyme levels. Brain hemorrhage was also seen in a female dog treated with atorvastatin for 3 months (Herron *et al*., 2015). Cats are less competent in drug metabolism than dogs, because of a defect in liver enzyme of glucuronyl transferase, although the exact mechanism is not known (Elliott, 2005). Accordingly, the use of chemical drugs, especially for prolonged periods, is guarded in cats. Unfortunately there are no long-term studies to verify the safety and efficacy of any lipid-lowering agent. In the present study, clinically relevant dosages of atorvastatin and chitosan were well tolerated for a period of 45 days. At the end of study, all cats were healthy and no clinical side effects were found in them.

Few studies have been conducted to compare the efficacy of chitosan and atorvastatin in animal models. Cunningham *et al*. (2013) reported that atorvastatin was well tolerated and did not result in apparent adverse effects or biochemical abnormalities in healthy dogs. Briand *et al*. (2006a) announced that treatment with atorvastatin significantly decreased plasma total cholesterol, phospholipids and triglycerides in dogs. They stated that a high dose of atorvastatin increased HDL-apo A-I fractional catabolic rate in dogs. Another survey showed that atorvastatin prevents the effects of hypertension in beagle dogs (Chen *et al*., 2012). In the present study, the treated groups (B and C) had good results in lowering LDL, compared with group A, on day 45. In comparison between two drugs and their effects on HDL, a significant difference was seen between groups A and B, on day 45.

Obesity is one of the most common clinical problems in cats presenting to veterinary practitioners. Persistently elevated lipid concentrations in cats result in the development of lipemia retinalis, peripheral neuropathy, cutaneous xanthomatosis and less commonly, anemia. These clinical manifestations can be successfully managed by feeding a low-fat diet (Knight, 2005; Kluger *et al*., 2009). In our survey, the main reason for the resistance to the effects of cholesterol in induced hyperlipidemia was unclear in cats (two cases). It may be due to genetic or species resistance that needs more research.

Now many medications are provided for the treatment of hyperlipidemia and reduction of cardiovascular disorders in humans and different animal species. Some synthetic antihyperlipidemic components have shown toxic effects. Hence attention has been given to naturally occurring antihyperlipidemic components (Stalenhoef *et al*., 2000; Ito *et al*., 2009). Gemfibrozil has been used to stimulate lipoprotein lipase activity and decrease VLDL secretion in humans. It is commonly recommended in combination with dietary therapy when the latter fails to lower serum triglyceride concentration below 500 mg/dl (Bauer, 2004). Fish oil supplement has resulted in a marked decrease in serum triglyceride and cholesterol concentrations in humans, rats, chicks, dogs, and rabbits (Mazaki-Tovi *et al*., 2014). The addition of vitamin E to the fish oil therapy regimen may enhance beneficial effects by increasing glutathione reductase activity and decreasing peroxide levels (Puiggros *et al*., 2002). However, little is known about the effectiveness

of fish oil therapy in cats. Niacin is a vitamin that has been used successfully for the treatment of hyperlipidemia without causing any side effects in humans (Kashyap *et al*., 2002). Garlic extracts have been used to decrease cholesterol level, but their effectiveness has not been evaluated properly in cats (Yen, 2001). Thyroxin therapy can decrease serum total cholesterol, and is effective in lowering serum lipid concentrations in hypothyroid dogs, but its use has not been recommended for cats (Ito *et al*., 2009). Periodic retesting of serum lipid concentrations should be re-evaluated for about 4-8 weeks after the treatment (Stalenhoef *et al*., 2000; Serisier *et al*., 2008). Knowledge of hyperlipidemia as a risk factor for diseases can heighten awareness and target health screening of cats. With the evidence from canine research studies, practitioners may be able to manage hyperlipidemia or overweight prevention plans in cats. Since weight gain generally occurs slowly and insidiously, a prevention counseling may be prudent at the time of the spay or neuter surgery, especially for the animals prone to weight gain (Elliott, 2005). In conclusion, we evaluated the anti-hyperlipidemic activity of chitosan compared with atorvastatin in cats. It was shown that although both drugs had hypolipidemic activity, the maximum percentage reduction of triglyceride level was observed with atorvastatin treatment. This may due to the better ability of atorvastatin to increase lipoprotein lipase activity than chitosan. Further studies will be needed to illuminate the mechanisms of action and their relative influence on biological functions. These surveys should be focused on the spectrum of conditions associated with hyperlipidemia, determining the underlying cause of idiopathic and familial forms of hyperlipidemia, as well as conducting clinical trials on the efficacy and safety of lipid-lowering drugs. The results of such experimental studies can greatly benefit the management of cats with hyperlipidemia.

## Acknowledgement

We would like to mention our great appreciation to the Research Council of Shahid Chamran University of Ahvaz, for their financial support.

## Conflict of interest

None of the authors have any conflict of interest to declare.

## References

**Allen Last, MD; Jonathan, D and Julianne Falleroni, DO** (2011). Pharmacologic treatment of hyperlipidemia. Am. Fam. Physician. 84: 551-558.

**Bauer, JE** (2004). Lipoprotein-mediated transport of dietary and synthesized lipids and lipid abnormalities of dogs and cats. J. Am. Vet. Med. Assoc., 224: 668-675.

**Briand, F; Magot, T; Krempf, M; Nguyen, P and Ouguerram, K** (2006a). Effects of atorvastatin on high-density lipoprotein apolipoprotein A-I metabolism in dogs. Eur. J. Clin. Invest., 36: 224-230.

**Briand, F; Serisier, S; Krempf, M; Siliart, B; Magot, T; Ouguerram, K and Nguyen, P** (2006b). Atorvastatin increases intestinal cholesterol absorption in dogs. J. Nutr., 136: 2034-2036.

**Chen, D; Zhou, D; Qian, J; Chen, F; Guan, L; Dong, L and Ge, J** (2012). Atorvastatin prevents dehydromonocrotaline-induced pulmonary hypertension in beagles. Exp. Lung Res., 38: 333-343.

**Choi, CR; Kim, EK; Kim, YS; Je, JY; An, SH; Lee, JD; Wang, JH; Ki, SS; Jeon, BT; Moon, SH and Park, PJ** (2012). Chitooligosaccharides decreases plasma lipid levels in healthy men. Int. J. Food Sci. Nutr., 63: 103-106.

**Cunningham, SM; Rush, JE and Freeman, LM** (2013). Short-term effects of atorvastatin in normal dogs and dogs with congestive heart failure due to myxomatous mitral valve. J. Vet. Intern. Med., 27: 985-989.

**Elliott, DA** (2005). Dietary and medical considerations in hyperlipidemia. In: Ettinger, SJ and Feldman, EC (Eds.), *Textbook of veterinary internal medicine*. (6th Edn.), St. Louis, Missouri, Saunders Elsevier. PP: 592-595.

**Ford, RB** (1996). Clinical management of lipemic patients. Compend. Contin. Educ. Vet., 18: 1053-1060.

**Herron, CE; Brueckner, CC; Chism, JP; Kemp, DC; Prescott, JS; Smith, GA; Melich, DH; Oleas, N and Polli, JW** (2015). Toxicokinetics and toxicity of atorvastatin in dogs. Toxicol. Appl. Pharmacol., 289: 117-123.

**Ito, BR; Zhang, BH; Cable, EE; Song, X; Fujitaki, JM; MacKenna, DA; Wilker, CE; Chi, B; van Poelje, PD; Linemeyer, DL and Erion, MD** (2009). Thyroid hormone beta receptor activation has additive cholesterol lowering activity in combination with atorvastatin in rabbits, dogs and monkeys. Br. J. Pharmacol., 156: 454-465.

**Johnson, MC** (2005). Hyperlipidemia disorders in dogs. Compend. Contin. Educ. Vet., 27: 361-364.

**Ju, C; Yue, W; Yang, Z; Zhang, Q; Yang, X; Liu, Z and Zhang, F** (2010). Antidiabetic effect and mechanism of chitooligosaccharides. Biol. Pharm. Bull., 33: 1511-1516.

**Kashyap, ML; McGovern, ME; Berra, K; Guyton, JR; Kwiterovich, PO; Harper, WL; Toth, PD; Favrot, LK; Kerzner, B; Nash, SD; Bays, HE and Simmons, PD** (2002). Long-term safety and efficacy of a once-daily niacin/lovastatin formulation for patients with dyslipi-demia. Am. J. Cardiol., 89: 672-678.

**Kluger, EK; Caslake, M; Baral, RM; Malik, R and Govendir, M** (2010). Preliminary post-prandial studies of Burmese cats with elevated triglyceride concentrations and/or presumed lipid aqueous. J. Feline Med. Surg., 12: 621-630.

**Kluger, EK; Hardman, C; Govendir, M; Baral, RM; Sullivan, DR; Snow, D and Malik, R** (2009). Triglyceride response following an oral fat tolerance test in Burmese cats, other pedigree cats and domestic crossbred cats. J. Feline Med. Surg., 11: 82-90.

**Knight, A** (2005). In defense of vegetarian cat food. J. Am. Vet. Med. Assoc., 226: 512-513.

**Kojuri, J; Vosoughi, AR and Akrami, M** (2007). Effects of anethum graveolens and garlic on lipid profile in hyperlipidemic patients. Lipids Health Dis., 6: 1-5.

**Maldonado, EN; Romero, JR; Ochoa, B and Aveldano, MI** (2001). Lipid and fatty acid composition of canine lipoproteins. Comp. Biochem. Physiol. B. Biochem. Mol. Biol., 128: 719-729.

**Mazaki-Tovi, M; Abood, SK and Schenck, PA** (2014). Fish oil supplementation increases concentration of adiponectin

in healthy dogs. J. Small Anim. Pract., 55: 247-253.

**Puiggros, C; Chacon, P; Armadans, LI; Clapes, J and Planas, M** (2002). Effects of oleic-rich and omega-3-rich diets on serum lipid pattern and lipid oxidation in mildly hypercholesterolemic patients. Clin. Nutr., 21: 79-87.

**Ross, CJ; Twisk, J; Bakker, AC; Miao, F; Verbart, D; Rip, J; Godbey, T; Dijkhuizen, P; Hermens, WT; Kastelein, JJ; Kuivenhoven, JA; Meulenberg, JM and Hayden, MR** (2006). Correction of feline lipoprotein lipase deficiency with adeno-associated virus serotype 1-mediated gene transfer of the lipoprotein lipase S447X beneficial mutation. Hum. Gene Ther., 17: 487-499.

**Sakamoto, S; Kashiki, M; Imai, N and Liang, CS** (1991). Effects of short-term, diet-induced hypercholesterolemia on systemic hemodynamics, myocardial blood flow, and infarct size in awake dogs with acute myocardial infarction. Circulation. 84: 378-386.

**Saravanan, G and Prakash, J** (2004). Effect of garlic (Allium sativum) on lipid peroxidation in experimental myocardial infarction in rats. J. Ethnopharmacol., 94: 155-158.

**Serisier, S; Gayet, C; Leray, V; Le Bloch, J; Ouguerram, K; Magot, T and Nguyen, P** (2008). Hyper-triglyceridaemic insulin-resistant obese dog model: effects of high-fat diet depending on age. J. Anim. Physiol. Anim. Nutr. (Berl)., 92: 419-425.

**Stalenhoef, AF; de Graaf, J; Wittekoek, ME; Bredie, SJ; Demacker, PN and Kastelein, JJ** (2000). The effect of concentrated n-3 fatty acids versus gemfibrozil on plasma lipoproteins, low density lipoprotein heterogeneity and oxidizability in patients with hypertriglyceridemia. Atherosclerosis. 153: 129-138.

**Tuccori, M; Montagnani, S; Mantarro, S; Capogrosso-Sansone, A; Ruggiero, E; Saporiti, A; Antonioli, L; Fornai, M and Blandizzi, C** (2014). Neuropsychiatric adverse events associated with statins: epidemiology, pathophysiology, prevention and management. CNS Drugs. 28: 249-272.

**Verd, JC; Peris, C; Alegret, M; Diaz, C; Hernandez, G; Vazquez, M; Adzet, T; Laguna, JC and Sanchez, RM** (1999). Different effect of simvastatin and atorvastatin on key enzymes involved in VLDL synthesis and catabolism in high fat/cholesterol fed rabbits. Br. J. Pharmacol., 127: 1479-1485.

**Xenoulis, PG and Steiner, JM** (2010). Lipid metabolism and hyperlipidemia in dogs. Vet. J., 183: 12-21.

**Xing, R; Liu, S; Guo, Z; Yu, H; Wang, P; Li, C; Li, Z and Li, P** (2005). Relevance of molecular weight of chitosan and its derivatives and their antioxidant activities *in vitro*. Bioorg. Med. Chem., 13: 1573-1577.

**Yen, YY and Liu, L** (2001). Cholesterol lowering effect of garlic extracts and organo-sulpur compounds: human and animal studies. J. Nut., 131: 989-993.

**Younes, I and Rinaudo, M** (2015). Chitin and chitosan preparation from marine sources. Structure, properties and applications. Mar. Drugs. 13: 1133-1174.