Effect of an oral dietary supplementation by a formulation based on silibinin-phosphatidylcholine complex in cats with liver diseases

Elena Biasibetti [1], **Tiziana Cocca** [2], **Maria Teresa Capucchio** [1], **Stefano Togni** [3], **Luca Giacomelli** [Corresp.], **Leonardo Giraudo** [4], **Mauro Bigliati** [4], **Marco Barbara** [5]

[1] Department of Veterinary Sciences, University of Turin, Grugliasco, Italy
[2] Clinica Veterinaria Napolivet
[3] Indena SpA
[4] Candioli farmaceutici
[5] University of Palermo, Italy

Corresponding Author: Luca Giacomelli
Email address: lu.giacomelli6@gmail.com

Background: Liver pathology in cats represents a frequent clinical condition occurring in animals of any age. Dietary supplementation with hepatoprotective and antioxidant products could represent a beneficial strategy to prevent and treat these disorders. We evaluated the tolerability and efficacy of long-term treatment with a multi-component, dietary supplement containing functional natural ingredients with recognized hepatoprotective properties in cats suffering from liver diseases
Methods: 20 European domestic cats of either gender, 10 with hepatic lipidosis and 10 with cholangitis received for 180 consecutive days a multi-component formulation based on the silibinin-phosphatidylcholine complex as paste or tablet. Clinical assessment and blood and urine parameters were evaluated at T0 (inclusion time) and after 7, 28, 60, 90 and 180 days from the initiation of the study.
Results: The oral supplementation with the silibinin-phosphatidylcholine complex significantly reduced the activity of liver enzymes in serum (alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase, γ -glutamyl transferase) and the content of total bilirubin, albumin, pre- and post- bile acids, in cats with hepatic lipidosis or cholangitis, normalizing their liver function parameters. Moreover, following the supplementation, the appetite was restored in all cats and the frequent episodes of diarrhea and vomit reported at inclusion almost disappeared.
Discussion: Dietary supplementation with the silibinin-phosphatidylcholine complex containing functional natural ingredients with recognized hepatoprotective and antioxidant actions might represent an effective strategy to improve the conditions of liver dysfunction, such as hepatitis lipidosis and cholangitis, in cats.

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1. Department of Veterinary Science, University of Torino, Torino, Italy
2. Direttore sanitario, Clinica Veterinaria Napolivet, Napoli, Italy
3. Business & cosmetics developments, Indena S.p.A, Milano, Italy
4. Department of Surgical Sciences and Integrated Diagnostics, School of Medicine, University of Genova, Genova, Italy
5. Regulatory affairs officer, Candioli Farmaceutici S.p.A, Torino, Italy
6. Responsabile scientifico, Candioli Farmaceutici S.p.A, Torino, Italy
7. Dipartimento Biomedico di Medicina Interna e Specialistica, University of Palermo, Palermo, Italy

***Corresponding author**

Luca Giacomelli, PhD

Department of Surgical Sciences and Integrated Diagnostics,

School of Medicine, Genova University

Genoa, Italy

Email: lu.giacomelli6@gmail.com

Abstract

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Results: The oral supplementation with the silibinin-phosphatidylcholine complex significantly reduced the activity of liver enzymes in serum (alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase, γ -glutamyl transferase) and the content of total bilirubin, albumin, pre- and post- bile acids, in cats with hepatic lipidosis or cholangitis, normalizing their liver function parameters. Moreover, following the supplementation, the appetite was restored in all cats and the frequent episodes of diarrhea and vomit reported at inclusion almost disappeared.

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Keywords: silibinin-phosphatidylcholine complex, cat, hepatic lipidosis, cholangitis

Introduction

Common hepatic diseases in small animals, including acute or chronic hepatitis, vascular anomalies, toxic liver diseases, hepatic lipidosis, and neoplasia, present a wide range of clinical signs, according with the nature and severity of disease. Lethargy, vomiting, diarrhea, and hyporexia are frequent early signs of liver diseases whereas hypoglycemia, petechiae, melena, and hematochezia are prevalent signs of advanced liver disease with decreased functional liver mass (Norton et al., 2016). Icterus, polyuria, and polydipsia can be observed at any stage, depending on the etiology of the liver disease (Norton et al., 2016). The diagnosis and treatment of canine and feline liver diseases have dramatically improved over the last 20 years due to the growing interest in hepatic histology and the hard work of small animal pathologists and clinicians to obtain a definitive diagnosis. In 2006, the World Small Animal Veterinary Association (WSAVA) Liver Standardization Group proposed common guidelines for diagnosis of liver diseases in dogs and cats, using both histological and clinical criteria (Rothuizen J, et al., 2006). A retrospective Japanese study applying the WSAVA diagnostic criteria showed that the most frequent canine liver disease was microvascular dysplasia (MVD) (29.4%), followed by hepatitis (23.5%) including cholangiohepatitis and cholangitis (Hirose et al., 2014). On the other

hand, the most frequent feline hepatic disease was parenchymal and interstitial hepatitis (45.1%), including neutrophilic cholangiohepatitis, lymphocytic cholangiohepatitis and neutrophilic cholangitis. The second most prevalent non-proliferative liver disease in cats was hepatic degeneration (14.1%), including lipidosis.

In particular, cholangitis/cholangio-hepatitis are acquired inflammatory liver disease classified as acute and chronic neutrophilic cholangitis (NC), lymphocytic cholangitis (LC), and chronic cholangitis associated with liver fluke infestation (CC), based on the composition of the infiltrate (Callahan et al., 2011). Hepatic lipidosis, also known as fatty liver syndrome, is a degenerative process characterized by the abnormal accumulation of lipid vacuoles in hepatocytes, leading to icterus, anorexia, and dramatic weight loss (Center et al., 1993). In all serious liver disorders, blood analysis may reveal elevated activities of hepatic enzymes, such as alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP), γ-glutamyl transferase (GGT), and increased levels of bile acids (Center et al., 1993). As liver functions decrease, the risk for presence of free radicals and oxidative injury increases. Several dietary supplements such as vitamins, L-carnitine, curcumin, and silymarin (extract of milk thistle), through their antioxidant action, could prevent liver damage and promote hepatocellular repair (Norton et al., 2016). In this study, we evaluated the tolerability and efficacy of long-term treatment with a multi-component, dietary supplement containing functional natural ingredients with recognized hepatoprotective properties in cats suffering from liver diseases.

Methods

Study design

20 European domestic cats of either gender, 10 with hepatic lipidosis and 10 with cholangitis were included in this study. The diagnosis of hepatic disease was based on clinical and radiographic signs and liver biopsy. Criteria for exclusion were: concomitant metabolic diseases or disorders that could impact liver functions (such as diabetes mellitus, gastritis, inflammatory bowel disease, chronic kidney disease, Cushing syndrome); cats treated with specific hepatoprotective products in the 30 days before inclusion; cats treated with antibiotics or anti-inflammatory drugs in the 2 weeks before inclusion. Owners were verbally informed about the methods and objectives of the study, and had to provide consent before their cat was enrolled.

Blood parameters and urine analysis as well as clinical examination were performed at inclusion time (T0) and after 7 (T7), 28 (T28), 60 (T60), 90 (T90) and 180 (T180) days from the initiation of the study.

Treatment

The treatment consisted of GlutaMax® FORTE paste or tablets. The paste was administrated up to 28 days, subsequently, from days 29 to the end of experiment, the multi-component formulation was administrated in form of tablets. 1 ml of paste per 3.5 kg body weight or 1 tablet per 10 kg body weight were daily administrated directly into the mouth or mixed with food for 180 consecutive days. In case of severe anorexia, the paste was mixed with liquid food and administrated through a nasogastric feeding tube.

The composition of the paste and tablets is reported in Table 1 and 2.

Statistical analysis

Data were summarized using descriptive statistics, mean and standard deviation were reported for continuous variables. Continuous variables were analyzed by using a mixed-effect linear model. In an ancillary analysis, markers of hepatic function in the treated cats were compared

with those reported in a historical cohort of cats with either lipidosis (n=10) or cholangitis (n=10) which did not receive Glutamax® FORTE supplementation but only standard management. Comparisons were performed by the Student's t test. A P-value <0.05 was considered statistically significant.

Results

Changes in clinical, hematological, coagulative, biochemical, and urine parameters in cats with hepatic lipidosis, and cholangitis are shown in Table 3 and Table 4, respectively. Consistently with the liver pathologic conditions, the levels of bilirubin, bile acids and liver enzyme activity in serum (ALT, AST, ALP, GGT) were remarkably high at baseline. Moreover, both cats with hepatic lipidosis as well as those with cholangitis presented at inclusion a mild hyperglycemia and abnormal coagulation profile, particularly in the fibrinogen level. Following supplementation with the multi-component product based on silibinin-phosphatidylcholine complex, the activity of liver enzymes steadily decreased, starting from the seventh days of treatment until the end of the study (T180), in both pathological conditions (Figure 1, 2, 3 and 4). Also the other serum biochemical parameters tend to normalize following the oral supplementation.

Among cats with hepatic lipidosis nobody showed appetite at the beginning of the trial. Due to the severe anorexia 6/10 were fed through a nasogastric feeding tube. Moreover, at inclusion, 6/10 cats suffered from vomit and diarrhea episodes. Starting from T28 until the end of the study, no cats with hepatic lipidosis were fed through a nasogastric feeding tube and vomit and diarrhea episodes disappeared in all cats.

On the other hand, 80% of cats with cholangitis were fed by nasogastric feeding tube at inclusion and 90% suffered from diarrhea and vomit. Starting from T60, all cats with cholangitis received

food and supplementation without the nasogastric tube. The frequency of diarrhea and vomit episodes variably decreased during the study, and at T180 only 2 cats presented rare episodes. Moreover, body weight and body condition significantly improved in all cats with cholangitis (Table 4).

Ancillary analysis

At baseline, there were no difference in hepatic function between cats treated with Glutamax® FORTE and controls (Table 5). However, 180 days since the institution of treatment, cats receving Glutamax® FORTE experienced a more marked reduction in ALT, AST and alkaline phosphatase, with respect to those on standard management (Table 6).

Discussion

Several mechanisms can be involved in the pathogenesis of liver diseases. Despite advances in modern medicine, there is no successful therapeutic approach regarding stimulation of hepatic function, liver protection or enhancement of hepatic cell regeneration (Madrigal-Santillan et al., 2014). The use of natural products in the treatment of these diseases has a long history. These products emerged as a promising source of relatively nontoxic compounds whose molecular mechanisms of hepatoprotective activity is difficult to understood. In fact if several formulations are pure substances with standardized formulas, most of them contain numerous constituents, making it difficult to attribute biological activity and mechanism to a specific compound. Hepatoprotective effects of natural compounds have been frequently attributed to their antioxidant properties and the ability to mobilize endogenous antioxidant defense system. Because of involvement of oxidative stress in virtually all mechanisms of liver injury, it is a reasonable presumption that antioxidant properties of these compounds may play a key role in

the mechanism of their hepatoprotective activity. Nevertheless, growing evidence suggests that other pharmacological activities of natural compounds distinct from antioxidant are responsible for their therapeutic effects. Numerous hepatoprotective medications with antioxidant properties are available on the market, including silymarin and other flavonoids, vitamins C and E. In particular, silymarin is a standardized extract of *Silybum marianum* (milk thistle) fruits and seeds containing multiple flavonolignans and flavonoids, among which silibinin is the predominant and the most active compound; this flavonoid complex presents several beneficial actions useful in the treatment of hepatic disease (Hacket et al., 2013). Sylimarin showed a hepatoprotective action by reducing liver enzyme activity in experimentally-induced liver damages in several animal models (Paulova et al., 1990; Wang et al., 1996; Lieber et al., 2003; Avizeh et al., 2011). However, similar to other flavonoids, sylimarin is not well-absorbed; therefore, the sylimarin most active molecule, silibinin, was combined with phosphatidylcholine to increase its bioavailability (Kidd et al., 2005). The hepatoprotective and antifibrotic effects of a silibinin could be increased also by the addition of vitamin E–phospholipids as demonstrated by Di Sario and collegues in a rat model of chronic liver disease (Di Sario et al., 2005). The best known mechanism of action of sibilinin is the antioxidant free radical scavenging and inhibition of lipid peroxidation, but anti-inflammatory effects are caused by nuclear DNA/RNA-mediated effects, via suppression of tumor necrosis factor (TNF), and interleukin expression involved in hepatic apoptosis and acute hepatitis *in vitro* and *in vivo* with decrease in liver enzyme activity. Moreover silibinin inhibits expression of the adhesion molecule selectin, necessary for leukocyte migration (Hackett et al., 2013). An antifibrotic effect in the liver with limitation in conversion of hepatic stellate cells to myofibroblasts and limitation of fibrous tissue production was also demonstrated. Hepatoprotective effects of silibinin in liver diseases may be related to

mechanisms of enhanced protein synthesis too, resulting in a more efficient hepatic regeneration and repair after toxic and inflammatory insults (Hackett et al., 2013).

Since cats more than other small animals are particularly susceptible to liver damage, supplementation with silibinin-phosphatidylcholine complex could represent a beneficial strategy to prevent and treat liver pathologies.

In this study, a multi-component formulation based on silibinin-phosphatidylcholine complex was administrated as dietary supplementation in cats with spontaneous liver diseases. This oral supplementation remarkably and significantly decreased the activity of liver enzymes in serum and the content of total bilirubin, albumin, pre- and post- bile acids, in cats with hepatic lipidosis or cholangitis, normalizing their liver function parameters. Interestingly, a more evident decrease of some markers of hepatic function (ALT, AST, alkaline phosphatase) was reported in cats treated with the multi-component formulation, with respect to those on standard management only. Moreover, following the supplementation, the appetite was restored in all cats and the frequent episodes of diarrhea and vomit reported at inclusion almost disappeared. Beside silibinin-phosphatidylcholine complex, other ingredients of this multi-component formulation, such as vitamin E, curcumin, zinc and citrus polyphenol, possess anti-inflammatory and antioxidant properties and could be associated with the beneficial effects of this supplementation (Filburn et al., 2006; Lerav et al., 2011).

However, we are aware that our research presents some study design limitations such as the small sample size.

Conclusions

The multi-component formulation based on the silibinin-phosphatidylcholine complex, might represent an effective strategy to improve the condition of liver dysfunction, such as hepatitis

lipidosis and cholangitis, in cats. Further investigations are need to better evaluate the compounds of this multi-component formulation which exhibit a most important beneficial effect against liver diseases.

List of abbreviations

ALP = alkaline phosphatase; ALT = alanine aminotransferase; AST = aspartate aminotransferase; CC = chronic cholangitis; GGT = γ-glutamyl transferase; LC = lymphocytic cholangitis; MVD = microvascular dysplasia; NC = neutrophilic cholangitis; TNF = tumor necrosis factor; WSAVA = World Small Animal Veterinary Association.

Competing interests*:* LeG and MB are employees of Candioli Farmaceutici S.p.A. ST is employee of Indena S.p.A. LuG is a consultant of Indena S.p.A.

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304 **Figures**

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Figure 1(on next page)

Changes in Alkaline Phosphatase values at each time-points, in both study groups

Figure 1. Changes in Alkaline Phosphatase values at each time-points, in both study groups

Figure 1

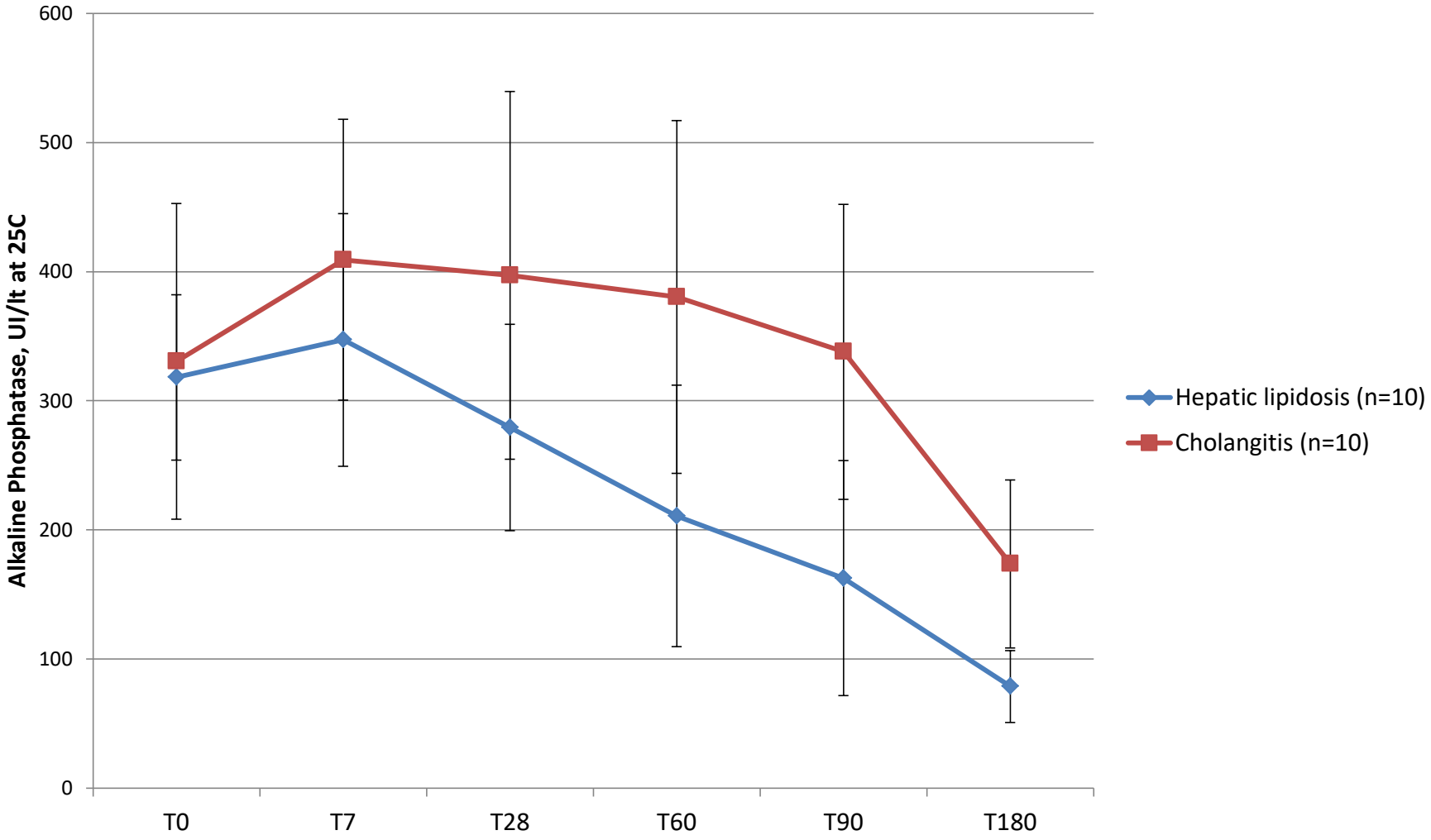

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Figure 2(on next page)

Changes in alanine aminotransferase values at each time-points, in both study groups

Figure 2. Changes in alanine aminotransferase values at each time-points, in both study groups

Figure 2

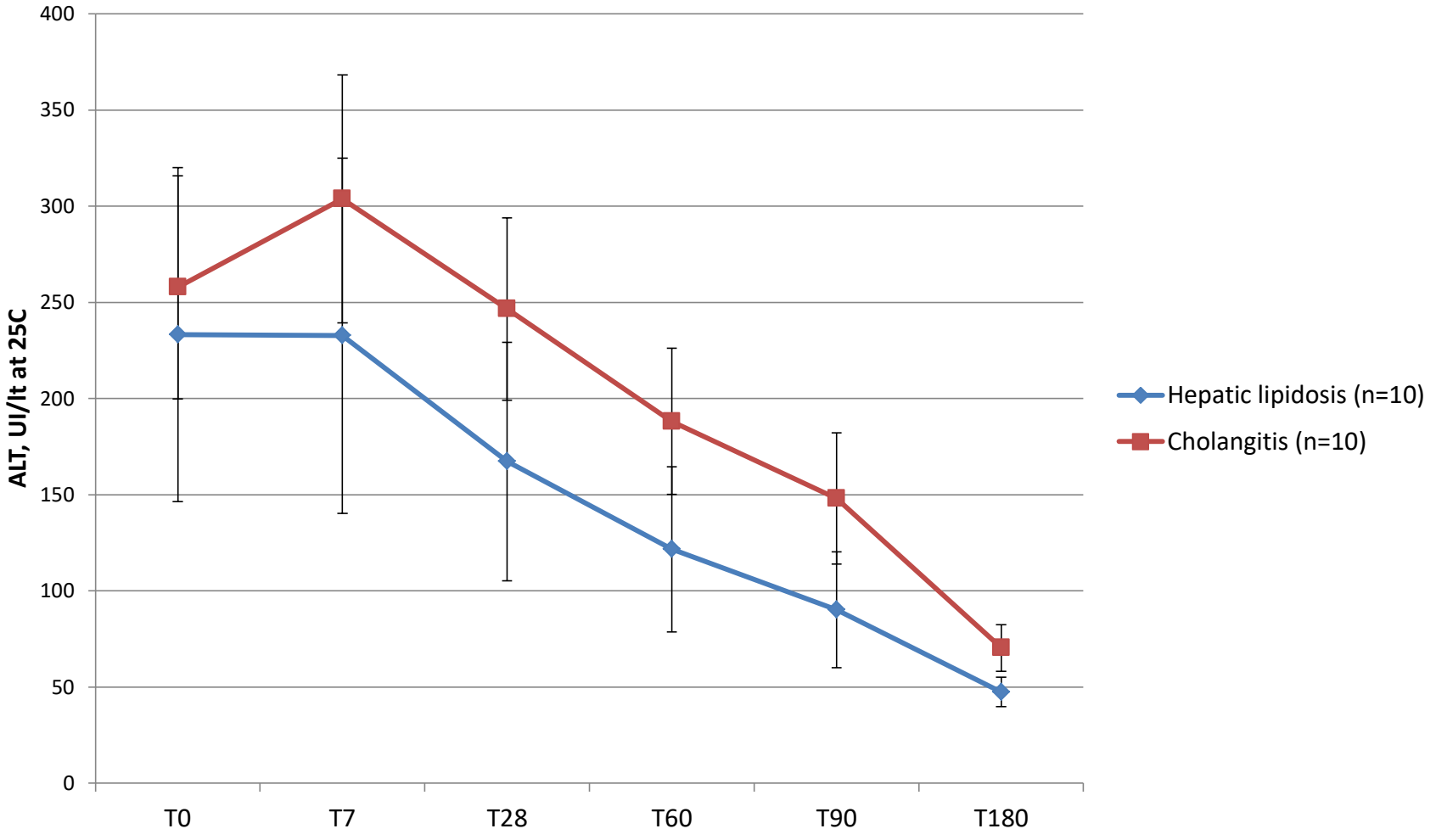

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Figure 3(on next page)

Changes in aspartate aminotransferase values at each time-points, in both study groups

Figure 3. Changes in aspartate aminotransferase values at each time-points, in both study groups

Figure 3

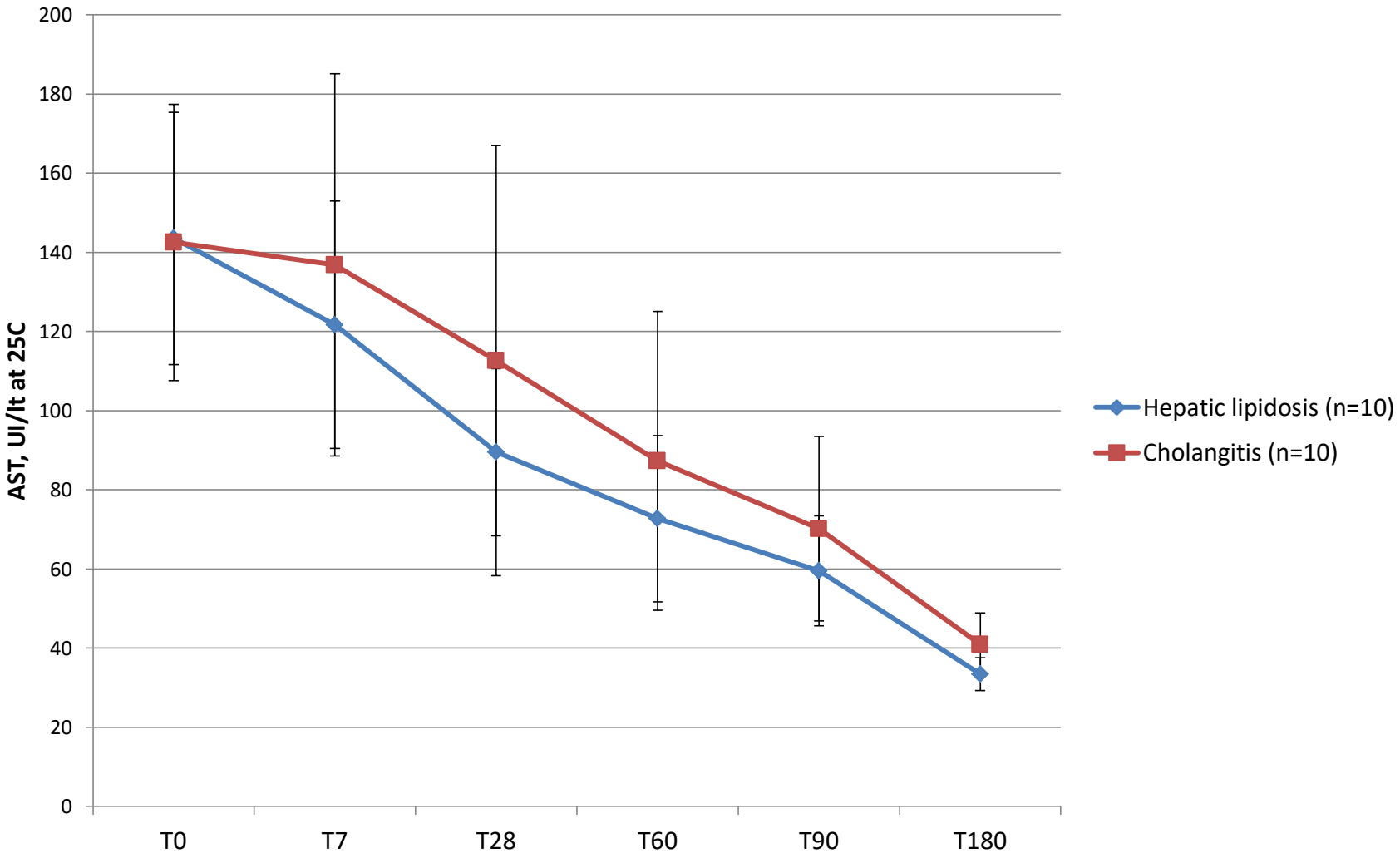

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Figure 4(on next page)

Changes in γ-glutamyl transferase values at each time-points, in both study groups

Figure 4. Changes in γ-glutamyl transferase values at each time-points, in both study groups

Figure 4

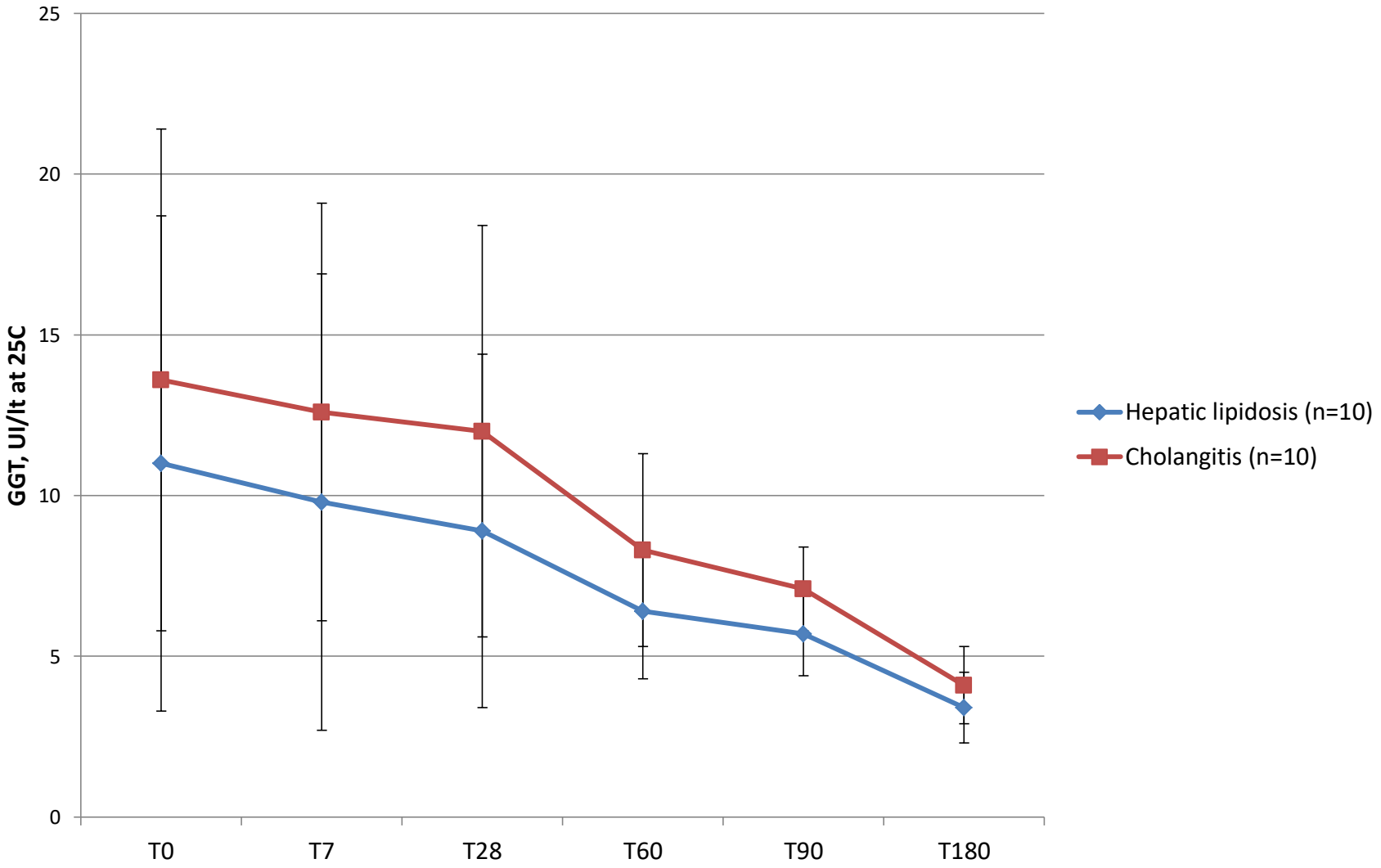

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Table 1(on next page)

Table 1

Table 1

1 Table 1. Composition of paste

Composition	**Additives (per kg)**	**Natural products**	**Elements**
Vegetable oil and fat (soybean oil, sunflower oil)	vitamin E/all-rac-alpha-tocopheryl acetate 3a700 mg 20,000	*Silybum marianum (L.) Gaertn.* (milk thistle extract CoE 551) mg 60,000	zinc oxide E 6 mg 300.
pig liver	L-carnitine mg 50,000	*Citrus aurantium L. var. dulcis* (orange peel extract CoE 143) mg 25,000	
malt extract	taurine mg 25,000	*Curcuma longa L.* (Turmeric extract CoE 163) mg 3,000	
glycerol or magnesium mono stearate	vitamin B12 mg 10		
maltodextrin			
Siliphos® (silybin/phospholipids) (Indena S.p.A., Milano)			

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Table 2(on next page)

Table 2

Table 2

1 Table 2. Composition of tablets

Composition	**Additives (per kg)**	**Natural products**	**Elements**	**Emulsifying and stabilizing agents**	**Binders, anti-caking agents and coagulants**
Pig liver	vitamin E/all-rac-alpha-tocopheryl acetate 3a700 mg 53,250	*Citrus aurantium L. var. dulcis* (orange peel extract CoE 143) mg 53,500,	zinc chelate of amino acids hydrate E6 mg 25,000.	microcrystalline cellulose E460 mg 261,000	colloidal silica E551b mg 17,400
dicalcium phosphate	choline bitartrate mg 17,450	*Curcuma longa L.* (Turmeric extract CoE 163) mg 8,830;			
mono and diglycerides of fatty acids (Behenic acid)	riboflavin mg 13,350				
magnesium stearate	thiamine hydrochloride 3a820 mg 4,250				
maltodextrin	pyridoxine hydrochlorid				

	e 3a831 mg 2,900				
Siliphos® (silybin/phospholipids) (Indena S.p.A., Milano)	vitamin B12 mg 21				

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Table 3(on next page)

Table 3

Table 3

Table 3. Changes in clinical, hematological, coagulative, serum biochemical and urine parameters in cats with hepatic lipidosis

	Hepatic lipidosis (n=10)						
	T0	**T7**	**T28**	**T60**	**T90**	**T180**	**P-value**
Body weight	6.7±2.3	6.5±2.0	6.4±1.9	6.4±1.8	6.5±1.7	6.4±1.6	0.09
BCS (range 0-5)	4.3±0.7	4.2±0.9	4.6±0.7	4.7±0.7	4.6±0.5	4.5±0.7	0.263
Temperature, °C	38.8±0.7	38.5±0.3	38.7±0.2	38.7±0.2	38.6±0.3	38.6±0.1	0.942
Complete blood count							
Hematocrit (HT), %	33.2±1.8	32.7±1.9	32.9±2.0	34.1±2.1	37±2.6	42±1.8	**<0.001**
Hemoglobin (HB), g/dl	11.1±0.8	11.3±0.8	11.4±1.1	12.0±0.9	13.4±1.0	15.6±0.8	**<0.001**
RBC, 10^6/mm3	7.3±1.0	7.3±0.8	7.4±0.8	7.5±0.8	7.9±0.4	8.7±0.3	**<0.001**
WBC, 10^3/mm3	24.5±4.0	22.8±3.2	15.8±1.2	12.8±0.9	10.7±1.6	9.0±1.2	**<0.001**
Platelets	467.1±89.7	467.3±74.7	401.5±77.2	433.3±105.5	389.1±71.9	334.2±50.5*	**<0.001**
Prothrombin Time (PT)	9.3±1.0	10.9±0.1*	8.5±0.9*	9.0±1.2*	8.7±1.0*	9.0±1.3*	0.475
Activated Partial Thromboplastin Time (APTT)	16.4±5.1	22.0±4.8*	15.8±2.2*	15.1±2.7*	14.9±1.6*	14.5±2.4*	ND
Fibrinogen, mg/dl	411.6±69.7	453.0±137.2*	369.6±68.8*	290.9±33.3*	276.8±73.8*	238.4±27.8*	**<0.001**
Serum biochemistry profile							
Blood Glucose, mg/dl	179.6±19.8	146.2±24.0	126.9±23.6	125.7±10.8	105.0±10.9	91.3±13.1	**<0.001**
BUN, mg/dl	61.1±14.3	57.7±12.1	48.5±11.5	41.6±7.1	40.1±5.2	36.9±5.8	**<0.001**
Creatinine, mg/dl	1.5±0.4	1.4±0.3	1.3±0.1	1.4±0.1	1.4±0.2	1.5±0.2	0.642
Total Protein, mg/dl	8.4±0.3	8.1±0.4	7.7±0.3	7.7±0.3	7.7±0.4	7.2±0.5	**<0.001**

Albumin, mg/dl	3.9±0.3	3.8±0.1	3.7±0.2	3.7±0.2	3.7±0.2	3.6±0.1	**0.001**
Albumin/Globulin, mg/dl	1.0±0.1	1.0±0.1	0.9±0.0	1.0±0.1	1.0±0.1	1.0±0.1	0.299
Total Bilirubin, mg/dl	2.6±0.7	2.1±0.6	1.1±0.4	0.7±0.3	0.5±0.2	0.3±0.1	**<0.001**
Alkaline Phosphatase, UI/lt a 25° C	318.1±64.1	347.1±97.9	279.3±80-0	210.8±101.4	162.6±91.1	78.7±27.8	**<0.001**
ALT, UI/lt at 25° C	233.1±86.8	232.6±92.3	167.3±62.0	121.6±43.0	90.1±30.1	47.4±7.6	**<0.001**
AST, UI/lt at 25° C	143.5±31.9	121.7±31.3	89.5±21.1	72.7±21.0	59.5±13.9	33.4±4.1	**<0.001**
GGT, UI/lt at 25° C	11.0±7.7	9.8±7.1	8.9±5.5	6.4±2.1	5.7±1.3	3.4±1.1	**<0.001**
Cholesterol, mg/dl	160.0±93.5	138.0±70.3	128.4±44.3	118.8±33.8	110.6±24.7	92.8±14.6	**<0.001**
Pre-meal bile acids	46.4±6.1*	39.7±10.0*	33.3±4.4*	28.4±3.4*	ND	12.9±5.7*	**<0.001**
Post-meal bile acids	ND	49.7±14.2*	41.0±3.8*	33.6±3.8*	ND	16.4±6.9*	**<0.001**
Urine test							
Urine Specific Gravity (USG)	1039.0±12.2	1034.0±9.3*	1031.0±6.3*	1037.0±7.0*	1037.0±10.4*	1036.0±6.3	0.964
Urine protein	226.9±84.2	182.0±46.3*	151.8±13.9*	147.9±28.8*	91.4±34.4*	90.0±38.7	**<0.001**

BCS: body condition score; RBC: red blood cells; WBC: white blood cells; BUN: blood urea nitrogen; ALT: alanine aminotransferase; AST: aspartate aminotransferase; GGT: γ -glutamyl transferase; ND: not determined; data are expressed as mean ± standard deviation; * data available for <10 cats

Table 4(on next page)

Table 4

Table 4

Table 4. Changes in clinical, hematological, coagulative, serum biochemical and urine parameters in cats with cholangitis

	Cholangitis (n=10)						
	T0	**T7**	**T28**	**T60**	**T90**	**T180**	**P-value**
Body weight, kg	3.4±0.4	3.2±0.4	3.5±0.3	3.6±0.4	3.8±0.3	4.2±0.3	**<0.001**
BCS (range 0-5)	2.4±0.5	2.2±0.4	2.5±0.5	2.5±0.5	2.7±0.5	3.0±0.0	**<0.001**
Temperature, °C	40.4±0.6*	39.2±0.4*	38.6±0.3*	38.6±0.3*	38.7±0.1*	38.6±0.3*	**<0.001**
Complete blood count							
Hematocrit (HT), %	37.1±14.7	34.6±12.3	34.0±8.4	37.5±7.2	38.0±4.8	43.3±2.4	ND
Hemoglobin (HB), g/dl	11.9±4.9	11.3±4.1	11.9±2.7	12.8±2.5	13.6±1.7	15.6±1.2	ND
RBC, 10^6/mm3	6.9±2.3	6.8±1.9	6.8±1.5	7.8±2.3	7.6±1.3	8.3±1.0	**<0.001**
WBC, 10^3/mm3	29.3±10.5	31.1±10.4	23.6±9.0	18.8±5.4	16.0±3.5	11.9±1.5	**<0.001**
Platelets	368.8±191.5	402.6±178.6	413.1±129.0	376.6±95.0	373.9±58.6	372.6±43.1	0.565
Prothrombin Time (PT)	9.8±2.2*	ND	9.6±2.7*	ND	9.6±1.3	ND	ND
Activated Partial Thromboplastin Time (APTT)	18.9±2.5*	ND	18.2±1.7*	ND	16.6±1.1	ND	ND
Fibrinogen, mg/dl	405.1±116.7*	ND	415.8±128.2*	ND	321.9±89.9	ND	ND
Serum biochemistry profile							
Blood Glucose, mg/dl	137.4±45.4	120.5±32.2	103.6±13.2	101.5±11.5	98.7±3.8	98.5±4.5	**0.001**
BUN, mg/dl	66.2±18.1	57.6±12.6	48.1±9.3	44.8±8.6	42.1±6.1	38.4±3.7	**<0.001**
Creatinine, mg/dl	1.6±0.5	1.6±0.4	1.5±0.3	1.4±0.2	1.4±0.2	1.6±0.2	0.789
Total Protein, mg/dl	8.1±1.1	7.8±1.1	7.6±1.0	7.5±0.7	7.6±0.5	7.7±0.2	0.3
Albumin, mg/dl	3.3±0.1	3.3±0.3	3.5±0.2	3.5±0.2	3.5±0.3	3.6±0.2	**<0.001**
Albumin/Globulin, mg/dl	1.0±0.3	1.0±0.3	1.0±0.2	1.0±0.2	1.0±0.1	0.9±0.1	0.355
Total Bilirubin, mg/dl	2.1±0.7	2.0±0.8	1.2±0.6	0.8±0.4	0.6±0.3	0.3±0.1	**<0.001**
Alkaline Phosphatase, UI/lt a 25° C	330.6±122.2	409.2±108.7	397.1±142.3	380.5±136.6	337.9±114.3	173.7±65.1	**<0.001**
ALT, UI/lt at 25° C	257.8±58.0	303.8±64.4	246.5±47.4	188.1±38.0	148.1±34.1	70.3±12.1	**<0.001**
AST, UI/lt at 25° C	142.5±34.9	136.8±48.3	112.6±54.3	87.3±37.8	70.2±23.3	40.9±8.0	**<0.001**
GGT, UI/lt at 25° C	13.6±7.8	12.6±6.5	12.0±6.4	8.3±3.0	7.1±1.3	4.1±1.2	**<0.001**
Cholesterol, mg/dl	84.7±64.1	85.0±49.3	101.0±34	108.1±24.5	116.8±21.1	89.9±12.0	0.479
Pre-meal bile acids	61.9±14.9	70.5±20.5*	42.7±12.4	ND	34.2±9.9	12.5±4.7*	**<0.001**
Post-meal bile acids	ND	91.5±17.7*	53.0±15.9	ND	41.6±12.1	21.1±7.4*	**<0.001**
Urine test							
Urine Specific Gravity (USG)	1042.0±13.4	1044.0±6.6*	1038.0±7.5	1040.0±3.5*	1036.0±8.2	1044.0±4.5*	0.541

Urine protein	175.2±47.9	184.1±67.6*	165.7±41.4	149.3±25.9*	139.0±32.8	111.4±22.3*	**<0.001**

BCS: body condition score; RBC: red blood cells; WBC: white blood cells; BUN: blood urea nitrogen; ALT: alanine aminotransferase; AST: aspartate aminotransferase; GGT: γ -glutamyl transferase; ND: not determined; data are expressed as mean ± standard deviation; * data available for <10 cats

Table 5(on next page)

Table 5

Table 5

Table 5. Markers of hepatic function of treated cats and controls at baseline. All data are expressed as mean (SD).

Lipidosis			
	Treated	*Control*	*p*
N	10	10	
Bun, mg/dl	61.1 (14.3)	61.50 (29.95)	0.970
ALT, UI/L	233.1 (86.8)	192.70 (77.02)	0.286
AST, UI/L	143.5 (31.9)	171.50 (113.20)	0.468
ALP, UI/L	318.1 (64.1)	259.00 (141.18)	0.250
GGT, UI/L	11.0 (7.7)	12.90 (5.67)	0.538
Cholangitis			
	Treated	*Control*	*p*
N	10	10	
Bun, mg/dl	66.24 (18.05)	52.20 (16.96)	0.090
ALT, UI/L	257.78 (58.01)	195.60 (87.27)	0.114
AST, UI/L	142.54 (34.87)	126.20 (55.20)	0.441
ALP, UI/L	330.63 (122.24)	220.10 (120.84)	0.057
GGT, UI/L	13.61 (7.78)	19.30 (7.96)	0.123

Table 6(on next page)

Table 6

Table 6

Table 6. Variations in markers of hepatic function of treated cats and controls after 180 days since institution of treatment. All data are expressed as mean (SD).

Lipidodis			
	Treated	*Control*	*p*
N	10	10	
Bun, mg/dl	-24.23 (11.56)	-20.20 (22.13)	0.618
ALT, UI/L	-185.66 (80.16)	-8.10 (90.06)	**<0.001**
AST, UI/L	-110.10 (30.10)	-37.60 (97.80)	**0.047**
ALP, UI/L	-239.43 (57.24)	-39.60 (141.91)	**0.001**
GGT, UI/L	-7.59 (8.08)	-4.10 (3.90)	0.241
Cholangitis			
	Treated	*Control*	*p*
N	10	10	
Bun, mg/dl	-27.83 (18.51)	-9.50 (16.95)	0.033
ALT, UI/L	-187.48 (55.30)	-49.00 (78.88)	**<0.001**
AST, UI/L	-101.64 (35.24)	-46.80 (49.59)	**0.011**
ALP, UI/L	-156.93 (76.47)	-16.50 (65.71)	**<0.001**
GGT, UI/L	-9.51 (8.30)	-9.10 (6.59)	0.904