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The role of Carcinine treatment on glico-lipidic imbalance of patients with altered blood glucose pattern

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Abstract

Objective. Several studies support the active role of Carcinine, an L-carnosine metabolite, in insulin resistance and dyslipidaemia, to modulate the insulinemic/glycaemic profile and fat metabolism.

Materials and methods. 100 (50 women and 50 men) volunteers, aged between 40 and 85 years with a body mass index (BMI) between 25 and 34,9 kg/m2, spontaneously addressed to our "Second Opinion Medical Consulting Network" (Modena, Italy) between 2019 and 2020, were included in this anecdotal, observational, retrospective trial. The aim of the study was to find an adequate possibly natural treatment for unbalanced insulin resistance pattern notwithstanding ongoing administration of statins, and hypoglycaemic chemical agents in healthy overweight/obese subjected. All the patients were divided in two groups: 1) the first group included 50 patients that were administered with a specific galenic nutraceutical product containing 20 mg of carcinine, and 2) the second group included 50 patients, which were administered with *lithothamnion calcareum* alga (190 mg) and three-time day for two months.

The waist circumference, glycaemia, homeostasis model assessment (HOMA-IR), glycated haemoglobin, total cholesterol values were detected at time 0, and time 1 (after treatment). At the same time, the pre versus post treatment, Advanced Glycation End products (AGEs), that play an important role in the development of diabetic vascular complications, were instrumentally measured at time 1 and 2.

Results. After 60 mg/day of Carcinina treatment, glycaemia (p=0,001), glycated haemoglobin (p<0,001), total cholesterol (p<0,003), serum insulin (p<0.05) were significantly reduced, respect to placebo period. Abdominal circumference (p<0.2), HOMA index (p<0.03) progressively were reduced as well. No cardiovascular risk and untoward effects were observed at the prescribed dosages. The AGE reader test showed a statistically meaningful reduced risk due to reduced amount.

Conclusions. Carcinine, at the daily dose of 60 mg/day, was able to modify, safely, the AGEs that induced cardiovascular risk, the waist circumference, and some glycolipid-metabolic parameters in overweight/obese patients with altered blood glucose pattern, improving significantly the impending metabolic syndrome. ***Clin Ter 2023; 174 (2):195-202 doi: 10.7417/CT.2023.2519***

Key words: *Carcinine, glycaemic state, lipid profile, AGEs*

Introduction

Metabolic Syndrome (MetS), also defined dysmetabolic syndrome or insulin resistance syndrome, is a clinical condition that includes multiple risk factors, such as obesity, hypertension, dyslipidaemia, and abnormal glucose metabolism associated with atherosclerotic cardiovascular disease (diabetes or stoke) (1). European MetS prevalence, using International Diabetes Federation diagnostic criteria, has been estimated as 41% in men and 38% in women (2). Several studies confirmed obesity as a "driving force" in the prevalence of insulin resistance and consequently of MetS, in combination with physical inactivity and atherogenic diet (3-6). In fact this nutritional excess can induct a hypoxic state, that can lead to cell necrosis with macrophage infiltration and production of adipocytokines ((interleukin-6, tumour necrosis factor α, as well as prothrombotix mediator plasminogen activator inhibitor 1 (PAI-1)) (7-10). Other potential mechanisms include low-grade chronic inflammation, oxidative stress and formation of advanced glycation end products (AGEs) (5, 11, 12). In fact, AGEs enclose a heterogeneous substance class (yellowish-brown materials with specific fluorescence), produced through non-enzymatic glycation of proteins, lipids or nucleic acids within the so-called Maillard reaction (MR) (13). Endogenous (e.g. oxidative stress, hyperglycaemia) and exogenous (e.g. cigarette smoke, high levels of refined and simple carbohydrate diets, hypercaloric diets, high temperature-cooked foods, and sedentary lifestyle) AGEs are partially degraded in the body or eliminated via the kidneys (13, 14).

In addition, AGEs bind to specific cellular receptors (REGEs), triggering a cascade of pro-inflammatory reactions. AGEs storage in the circulating blood and various tissues, contributes to the development of vascular complications in patients with and without diabetes, thus playing a central role in the long-term metabolic memory, called 'glycaemic memory' concept (13). However, AGEs represent the substrate of this glycaemic memory, and its level measurement might suggest the risk of diabetes complications than the HbA1c that mirrors glycaemic control over 8-12 weeks only (15, 16). The risk of diabetic complications by AGEs high levels

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can be reduced by natural antioxidant components: enclosing L-carnosine or Carcinine (Fig.1). The first is a naturally occurring dipeptide (β-alanyl-histidine), commonly found in the non-protein fraction of mammalian tissues, skeletal muscle, and brain (23, 24). It was first identified in 1900 in beef extract, and it is a potent endogenous scavenger of oxidative stress and highly concentrated (mM) in muscle and nervous tissues (17, 18). Instead, Carcinine (β-alanyl-L-histamine), a metabolite of L-carnosine, was discovered in cardiac tissue of the crustacean *Carcinus maenas* in 1973 and has since been identified in the hearts of other crustacea (19). Carcinine would be a better effective choice rather than carnosine because it is significantly more resistant to enzymatic hydrolysis than carnosine and has anti-glycation, anti-inflammatory and antioxidant properties, suggesting a potential strategy in MetS treatment (20-22).

The aim of our clinical retrospective observational trial was to evaluate the therapeutic action of carcinine on metabolic parameters in overweight/obese patients with altered blood glucose pattern.

Materials and methods

The study is a simple open investigation comparing two groups of patients with altered blood glucose pattern, applied between January 2019 and March 2020, to our "Second Opinion Medical Consulting Network" (Modena, Italy), looking for further nutritional supplements to improve life quality and counteract the symptoms of the diabetic and metabolic syndrome eligible for this anecdotal, spontaneous, and retrospective trial (23-26)**.** The Second Opinion Medical Network is a consultation referral web and Medical Office System enclosing a wide panel of specialists, to whom any patient with any illness or syndrome that is not adequately satisfied by the previous diagnosis or therapy can be applied for an individual telematic or front office clinical audit (27-30). Inclusion criteria: -partecipants at least 18 years of age, -healthy overweight/obese patients, -BMI range between 25 and 34,9 kg/m2, -altered blood glucose pattern, -patients with confirmed diagnosis of Type 1 or Type 2 diabetes mellitus for ≥1 year and using insulin by multiple-daily subcutaneous injections or insulin pump and an HbA1c > 8%, patients with insulin resistance defined as glycated haemoglobin (HbA1c) 5.5–6.4%, patients on metformin or simvastatin drug therapy.

Exclusion criteria: -patients with borderline psychiatric problems (anxiety, depression, insomnia) involving eating behaviour and lifestyle physical activity, with obvious rebound on metabolism, -subjects who did not accept to undergo biochemical exams, -pregnant women, -patients with clinical or laboratory signs of acute or chronic infection.

Recruitment criteria

100 Patients, aged between 40 and 85 years and with body mass index (BMI) between 25 and 34.9 kg/m2, with the plasma glucose level ≥126 mg/dl in fasting or ≥200 mg/dl for 2h value after 75 g glucose oral load, glycate haemoglobin ≥6.5 %, on metformin (500mg/day) and/or simvastatin (30mg/day) treatment (Table 1).

The 2 years average/ long standing daily intake of metformin 500 mg once a day and simvastatin 30 mg nighttime, was not given up even if inadequate, but integrated with experimental treatment. Any other drug (vitamins, benzodiazepines, or other hormonal thyroid) or antioxidant treatments were withdrawn 2 weeks before the start-up of the study. Standardization of the caloric amount to 1400 calories balancing carbohydrates 40%, proteins 30%, lipids 30% and daily physical activity (30 minutes walking every day) were alleged in the protocol. All the patients signed an informed consent and were followed weekly with phone interview and monthly with a clinical visit.

The study was conducted in accordance with the Declaration of Helsinki and approved by the local ethics committee Second Opinion Local Institutional Review Board (IRB). All the patients were divided in two groups comparable in terms of age, clinical history, symptoms: (1) the first group included 50 patients that were administered with a specific galenic nutraceutical product "Un-tafor" (3 cps/day before the main meals, breakfast, lunch, and dinner) for 2 months. Each capsule contains 200 mg of Glycoless (10% of carcinine (2HCl), β alanyl-histamine dihydrochloride), however 20 mg of carcinine; and (2) the second group included 50

Table 1. Baseline characteristics of the patients

Parameters	Values
Age (years)	58.2±20.3
Male (%)	50
Female (%)	50
Weight (kg)	85.4±15.2
BMI (kg/m2)	31.2±6.4
HbA1c (%)	8.7±2.5
Insulin resistance	9.5±7.2

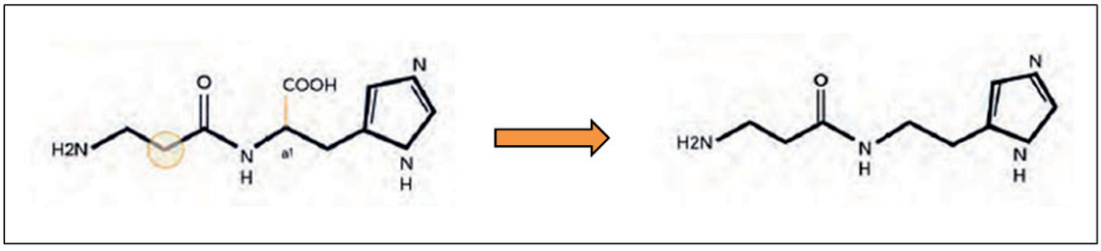

Fig. 1. Structural modification of the L-carnosine molecule converting into carcinine

patients, which were administered with a placebo, conventionally named "Un-tafor X", composed by *lithothamnion calcareum* alga (190 mg) and 10 mg of Glycoless (1 mg of carcinine) three-time day for two months.

At the end of this treatment, laboratory exams, (glycaemia, glycated haemoglobin, total cholesterol), waist circumference, and HOMA index were measured at time 1, and time 2 (after treatment).

HOMA-IR is a simple method described by Matthews et al., based on the hypothesis that basal glucose and insulin interactions are largely determined by a simple feedback loop (23).

Compliance to proposed lifestyle and variation in eating behaviour were also investigated in order to detect any influence of the newly added nutraceutical support compared with the old one on some nutrition and energy spending parameter. Also pre and post treatment, the level of Advanced Glycation End products (AGEs), that plays an important role in the development of diabetic vascular complications, was detected, using a point of care non-invasive monitoring device (AGE Reader, Diagnoptics Technologies B.V, The Netherlands).

Blood collection and measurement of biochemical indices

Blood samples were collected using the standard phlebotomy technique in the morning after a 12-hour overnight fast. The patients were registered at the front desk and sent for collection. Serum samples were rested for 20 minutes for clot retraction after centrifugation. The samples were centrifuged for 8 minutes at a speed of 3000 rpm. For lipidogram, glucose, CRP and uric acid, the VITROS 5, 1 FUSION automatic dispenser was used by the Dry Chemistry method. For fructosamine, the method Colorimetric was used; for VHS, the Westergren method; Insulin and ferritin was measured with chemiluminescence methodology; the high-performance liquid chromatography method was used for glycated haemoglobin measurement.

AGEs level measurement

Circulating or tissue-bound AGEs can be measured using enzyme linked immunosorbent assay (ELISA), fluorescence spectroscopy, fluid chromatography and gas chromatography with mass spectrometry (24). We selected an easy non-invasive method based on AGEs-related skin autofluorescence (AGE Reader, DiagnOptics, Groningen, the Netherlands) (25). The patient placed his forearm over the scanner on the blue field, so that no outside light interferes the 'measurement window' (Fig. 2 A-D). An excitation, autofluorescent light source (composed by multiple light emitting diodes (LEDs)) is directed through this window onto the skin for fractions of a second. This skin autofluorescence (SAF) is displayed on the screen within seconds, together with its age-adjusted reference values and a software specifies the risk level of endogenous AGEs pool (Fig. 3 A,B) (26).

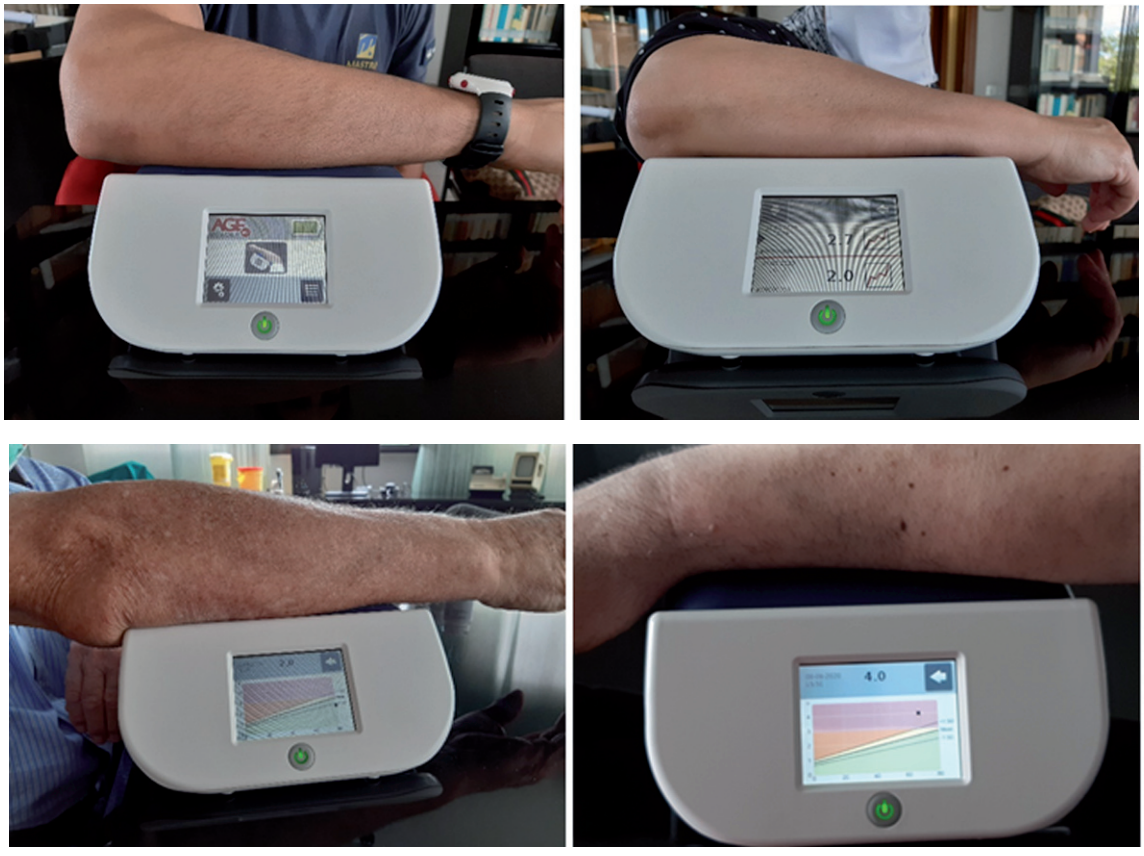

Fig. 2 (A-D): Position of the patient arm in the AGE reader

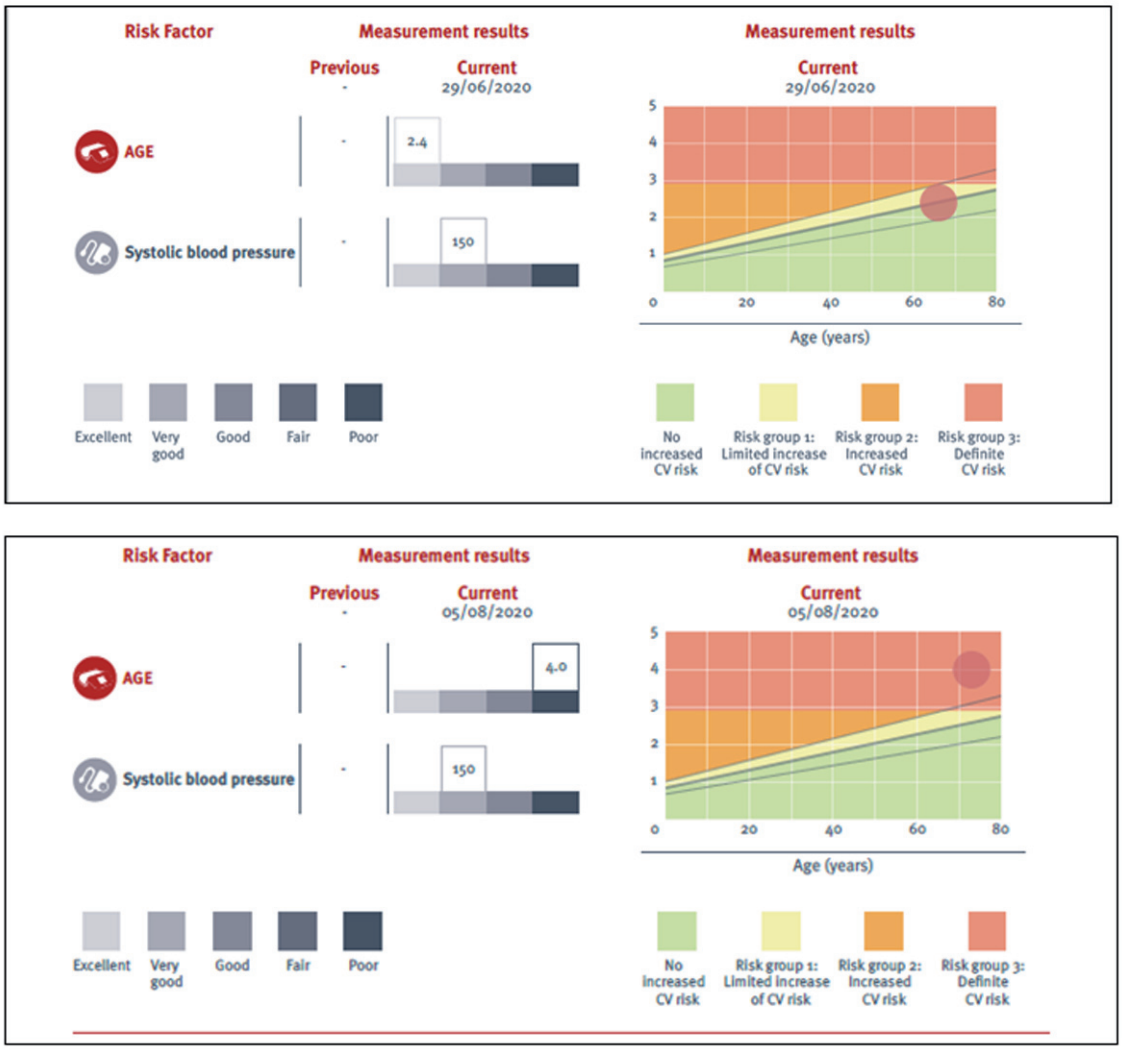

Fig. 3 (A,B): Graphical representation of results of AGE Reader

Statistical analysis

Statistical analysis was performed using GraphPad Prism 7 (GraphPad Software Inc., San Diego, CA, USA). Data were analyzed using an unpaired t-test with Welch's correction. $p < 0.05$ was considered significant.

Results

High dose Carcinine supplementation (first group) compared to "*lithothamnion calcareum* algae + carcinine" treatment (second group), significantly decreased glycemia ($p = 0,0412$) glycated haemoglobin ($p=0,0335$), total cholesterol ($p=0,0303$) and serum insulin ($p=0,0064$) after in all the patients. A significant reduction in abdominal circumference ($p <0.2$) and HOMA-IR ($p <0.03$) between time 1 and 2 was also observed (Table 2, Fig. 4-7).

Table 2. Biochemical parameters at time 1 (after carcinine treatment) and 2 (lithothamnion calcareum + carcinine treatment). Values are presented as mean±standard deviation.

Variable	Values at T1 (Carcinine treat.)	Values at T2 (lithothamnion calcareum algae + carcinine treat.)
Glycemia (mg/dL)	67.4±8.2	87.6±8.8
Glycated Haemoglobin (%)	7.4±1.2	8.5±1.3
Total Cholesterol (mg/dL)	181±24.4	190±21.4
Serum insulin (µU/mL)	12.3±7.6	17.2±8.0
Abdominal Circumference (cm)	95.84±7.00	98.70±5.00
HOMA-IR	2.9±1.5	4.1±1.3

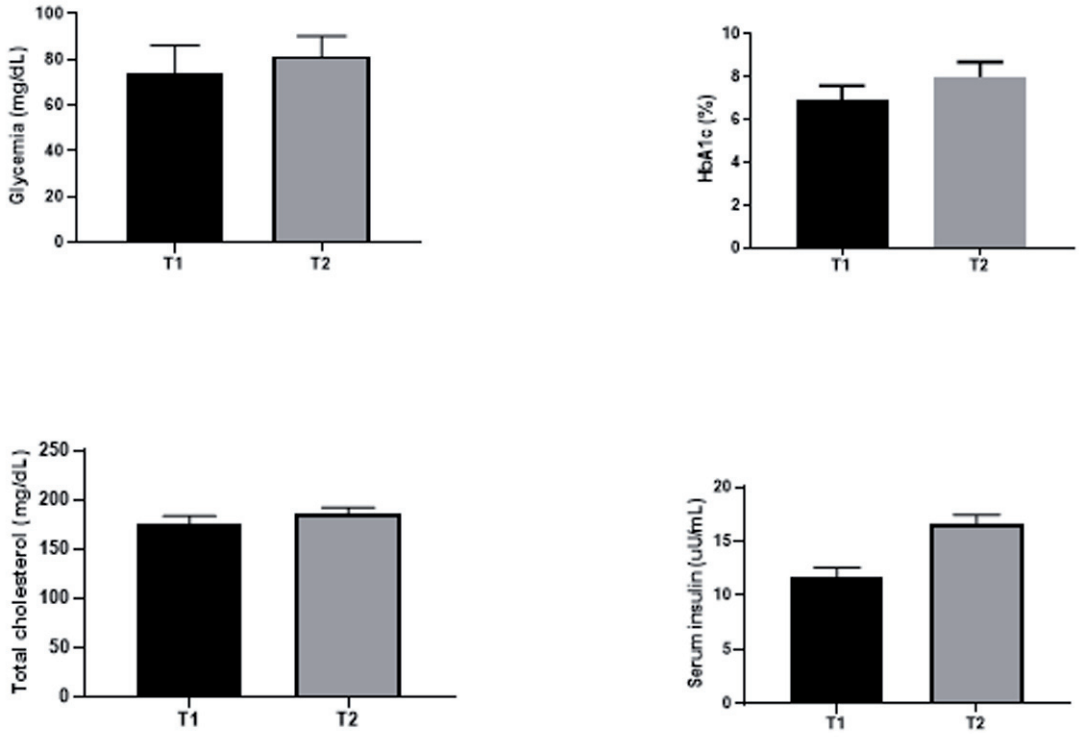

Fig. 4-7: Graphical representations of biochemical parameters at T1 and T2

The SAF showed a linear increase of value/year for persons up to 70 years, according to Koetsier et al.(31). Furthermore, we observed a reduced AGEs values after 60 days, with reduction of cardiovascular risk probability (Table 3).

There are several limitations of SAF measurements, such as the possibility that some external factors (e.g. creams, sunscreens, self-browning creams, extreme hyperaemia, or vasoconstriction, etc.) can influence SAF measurements as reported by Noordzij et al. (32). We carefully standardized the procedure in order to avoid these limits as much as possible.

Discussion

The potential action of L-carnosine as a pharmacological agent has been studied in rodent models of metabolic syndrome and cardiovascular disease (33-35). However, the bioavailability of L-carnosine as an oral drug therapy has several limitations, mainly the high serum and tissue carnosinase activity in humans degrades carnosine by rapid hydrolysis of the peptide bond (17). Carcinine, instead, exists in multiple-histamine-rich mammalian tissues such as heart, kidney, stomach and intestine (36). In particular, our study has been focused on the clinical benefits of carcinine administration, whose action mechanism can be better highlighted considering the chemical affinity of this molecule with carnosine whose physiological role has been extensively investigated (37, 38). A number of putative roles have been ascribed to the family of carnosine, such as intracellular buffering (39), regulation of glycogenolysis (40), muscle calpains and myosin activation, neurotransmission (41). Carnosine also provides of a weak antinflammatory tissue repair effect, especially in oral surgery (42). Experimentally, it protected hearth reperfusion injury after ischemia in the rabbit model (43, 44), and prevented, or partly reversed, lens cataract (45). Carnosine and carcinine appear to be physiological antioxidants able to efficiently protect the lipid phase of biological membranes and aqueous environments. Carnosine and carcinine exhibit an ability to inhibit lipid oxidase with scavenger activity and block of lipid peroxidation generation, they display antioxidant properties, chelating transition metals (21, 46). Differently from carnosine, being carcinine refractory to the enzyme carnosinase inactivation, it longer maintains and enhances the antioxidant effect of carnosine (20). The simple decarboxylation of carnosine to produce carcinine, creates a great difference in the catabolism of the 2 molecules. The inactivation of carcinine with arylamidase is 30 times weaker than that of carnosine, thus the former has a greater bioavailability than the latter. The most appealing antioxidant

Table 3. SAF values at T1 and T2

Age Group (years)	SAF value at T1 (Carcinine treat.)	SAF value at T2 (Lithothamnion algae+ carcin treat.)
40-55	2.2±1.4	2.3±1.7
55-70	3.7±2.5	3.9±3.1
70-85	3.8±2.7	4.0±3.2

clinical effects of this compound are potentially antiaging, preventing the reticulation damages of collagenic and elastic structures and also counteracting deoxyribose degradation by toxic aldehydes and ROS (47). Our study was addressed to explore the putative therapeutic role of carcinine to relieve the glycation products side effects and to reduce the risk factors due to the metabolic syndrome, a fascinating investigation area to prevent the cardiovascular damages of the glycation products induced by insulin resistance. We showed that 8 week substantial dosages of carcinine supplementation led to a significant decrease in fasting glycemia ($p = 0.001$). Similar results were obtained by Houjeghani (48), after 12 weeks of L-carnosine supplementation. The authors observed a significant reduction in serum glucose and glycated haemoglobin levels, but with no significant effect on HOMA-IR. De Courten published that patients with metabolic syndrome, and glucose intolerance supplemented for 12 weeks with L-carnosine, reduced the two-hour glucose and insulin levels without effects on fasting glycemia (49) .Yilmaz (50) in previous experimental trials, observed significant reductions in plasma glucose in animal models. Other in *vivo* studies described L-carnosine induced hypoglycaemia. Soliman (51) in streptozotocin-induced diabetic models showed that treatment with L-carnosine (100 and 200 mg/kg) improved glycaemic and dose-dependent dyslipidaemia parameters. Another study pointed out that oral supplementation with L-carnosine for 4 weeks reduced plasma glucose in animal models (52). The possible mechanisms by which carnosine leads to glucose reduction can be explained by Nagai (53) who demonstrated that carnosine increases parasympathetic nerves function in the pancreas, with subsequent stimulation of insulin secretion and reduced glucagon secretion from the pancreas, with an hypoglycaemic effect. Carnosine decreases, however, plasma glucose levels by inhibiting gluconeogenesis, increasing glycolysis and glucose sensitivity, as suggested by Tsoi (53). In our study, we detected a significant reduction in glycated haemoglobin ($p < 0.001$) in the 8 weeks administration schedule as reported in the meta-analysis of Menon (54). Similarly, Elbarbary (55) observed a reduction in glycated haemoglobin values ($p < 0.05$) in diabetic children supplemented with 12 weeks. Carnosine administration L-carnosine is able to inhibit the proteins glycation (56, 57). The abdominal circumferences were reduced as well at the end of 8 weeks-carcinine treatment ($p < 0.2$), in contrast with Houjeghani and De Courten who denied significant differences in abdominal circumference (48, 49). Abdominal circumference and visceral fat are considered an indicator of fatal risk and aspecific symptoms of the metabolic syndrome (20, 58). Its reduction reversibly decreases risk of the metabolic syndrome (59, 60). Reduction in total cholesterol levels ($p < 0.003$) at the end of 8 weeks was observed after carcinine supplementation, respect to placebo treatment. Likewise, Mong (61) observed in his study that carnosine supplementation in mice reduced total cholesterol levels in eight weeks. According to the same authors, such reduction was due to the decrease in the activity of cholesterol-regulating enzymes such as 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase. It is known that altered values of total cholesterol are associated with the metabolic syndrome (62). Thus, reducing total cholesterol levels decreases the risk factors associated with the syndrome. We found that Carcinine (L-carnosine metabolite) decreased some of the parameters of the metabolic syndrome in 8 weeks, respect to placebo treatment, thus indicating that the human model also has the same result as the animal model. The limitations of our study are: 1) small patient cohort, 2) absence of control group, 3) the use of HOMA-IR as single marker, since this is limited especially in patients with severe hyperglycaemia or non-obese diabetic patients, whose basal insulin secretion is impaired (63). Based on the results of our research and the findings from the studies mentioned above, carcinine supplementation appears to be an adjunct approach to improving fasting blood glucose, total cholesterol, insulin, HOMA-IR and abdominal circumference as metabolic markers and of glycated haemoglobin as markers of glycation.

Conclusions

In conclusion, 8 weeks of carcinine oral supplementation (60 mg/day) resulted in a decrease of glycated haemoglobin, total cholesterol, insulin, abdominal circumference, and HOMA-IR.

Notably from the symptomatic point of view 80% of the treated subjects reported after 2 weeks carcinine administration, a higher fasting threshold of appetite and reduced hunger before the meals.

Furthermore, quicker satiety after lunch and dinner without further required carbohydrate intake was detected, with reduced food amount and suppression of binge eating at day and night /time.

This very relevant clinical observations about eating, and lifestyle improvements might be explained with an enhanced insulin stability and balanced feedback release from the pancreatic insulae, without hypoglycaemic fasting post-prandial peaks.

This effect is worth in our opinion of specific carcinine prescription to reduce compulsive eating behaviour of metabolic syndrome.

Declaration of conflict of interest: The authors declare no conflict of interest with any financial organization regarding the material discussed in the manuscript.

References

1. Rochlani Y, et al. Metabolic syndrome: pathophysiology, management, and modulation by natural compounds. Therapeutic advances in cardiovascular disease, 2017; 11(8): 215-225
2. Gao W. Does the constellation of risk factors with and without abdominal adiposity associate with different cardiovascular mortality risk? Int J Obes (Lond), 2008; 32(5):757-62
3. Ginsberg HN. Insulin resistance and cardiovascular disease. J Clin Invest, 2000; 106(4):453-8
4. Reaven GM. Banting lecture 1988. Role of insulin resistance in human disease. Diabetes, 1988; 37(12):1595-607
5. Eckel RH, SM Grundy, PZ Zimmet, The metabolic syndrome. Lancet, 2005; 365(9468):1415-28

6. Grundy SM, et al. Diagnosis and management of the metabolic syndrome. An American Heart Association/National Heart, Lung, and Blood Institute Scientific Statement. Executive summary. Cardiol Rev, 2005; 13(6):322-7
7. Halberg N, I Wernstedt-Asterholm, PE Scherer, The Adipocyte as an Endocrine Cell. Endocrinology and Metabolism Clinics of North America, 2008; 37(3):753-768
8. Testa R, et al. Interleukin-6–174 G>C polymorphism affects the association between IL-6 plasma levels and insulin resistance in type 2 diabetic patients. Diabetes Research and Clinical Practice, 2006; 71(3):299-305
9. Azzawi M, P. Hasleton. Tumour necrosis factor alpha and the cardiovascular system: its role in cardiac allograft rejection and heart disease. Cardiovascular Research, 1999; 43(4): 850-859
10. Alessi MC, I Juhan-Vague, PAI-1 and the metabolic syndrome: links, causes, and consequences. Arterioscler Thromb Vasc Biol, 2006; 26(10):2200-7
11. Roberts CK, Hevener AL, Barnard RJ. Metabolic syndrome and insulin resistance: underlying causes and modification by exercise training. Comprehensive Physiology, 2013; 3(1): 1-58
12. Cheng C, et al. Expression profiling of endogenous secretory receptor for advanced glycation end products in human organs. Mod Pathol, 2005; 18(10):1385-96
13. Yamagishi S, Imaizumi T. Diabetic vascular complications: pathophysiology, biochemical basis and potential therapeutic strategy. Curr Pharm Des, 2005; 11(18):2279-99
14. Yamagishi S, Ueda S, Okuda S. Food-derived advanced glycation end products (AGEs): a novel therapeutic target for various disorders. Curr Pharm Des, 2007; 13(27):2832-6
15. Monnier VM, et al. Skin collagen glycation, glycoxidation, and crosslinking are lower in subjects with long-term intensive versus conventional therapy of type 1 diabetes: relevance of glycated collagen products versus HbA1c as markers of diabetic complications. DCCT Skin Collagen Ancillary Study Group. Diabetes Control and Complications Trial. Diabetes, 1999; 48(4):870-80
16. Cleary PA, et al. Clinical and technical factors associated with skin intrinsic fluorescence in subjects with type 1 diabetes from the Diabetes Control and Complications Trial/Epidemiology of Diabetes Interventions and Complications Study. Diabetes technology & therapeutics, 2013; 15(6):466-474
17. Boldyrev AA, Aldini G, Derave W. Physiology and pathophysiology of carnosine. Physiol Rev, 2013; 93(4):1803-45
18. Vishnyakova KS, et al. Stimulation of cell proliferation by carnosine: Cell and transcriptome approaches. Molecular Biology, 2014; 48(5):718-726
19. Fingerman M, et al. Biogenic Amines in Crustaceans: Identification, Localization, and Roles. Journal of Crustacean Biology, 1994; 14(3):413-437
20. Pegova A, Abe H, Boldyrev A. Hydrolysis of carnosine and related compounds by mammalian carnosinases. Comp Biochem Physiol B Biochem Mol Biol, 2000; 127(4):443-6
21. Babizhayev MA, et al. Skin beautification with oral non-hydrolized versions of carnosine and carcinine: Effective therapeutic management and cosmetic skincare solutions against oxidative glycation and free-radical production as a causal mechanism of diabetic complications and skin aging. J Dermatolog Treat, 2012; 23(5):345-84
22. Anderson EJ, et al. A carnosine analog mitigates metabolic disorders of obesity by reducing carbonyl stress. The Journal of Clinical Investigation, 2018; 128(12):5280-5293
23. Matthews DR, et al. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia, 1985; 28(7): 412-9
24. Mulder DJ, et al. Skin autofluorescence, a novel marker for glycemic and oxidative stress-derived advanced glycation endproducts: an overview of current clinical studies, evidence, and limitations. Diabetes Technol Ther, 2006; 8(5): 523-35
25. Meerwaldt R, et al. Simple non-invasive assessment of advanced glycation endproduct accumulation. Diabetologia, 2004; 47(7):1324-1330
26. Meerwaldt R, et al. Simple noninvasive measurement of skin autofluorescence. Ann N Y Acad Sci, 2005; 1043:290-8
27. Palmieri B, Iannitti T. The Web Babel syndrome. Patient Educ Couns, 2011; 85(2):331-3
28. Palmieri B, et al. (Second opinion clinic: is the Web Babel Syndrome treatable?). Clin Ter, 2011; 162(6):575-83
29. Palmieri B LC, Vadalà M. The "Second Opinion Medical Network". Int J Pathol Clin Res, 2017; 3(056)
30. Wunsch, A.a.B.P., The rôle of second opinion in oncology: an update. 2014. 18(3): p. 117-120.
31. Koetsier, M., et al., Reference values of skin autofluorescence. Diabetes Technol Ther, 2010. 12(5): p. 399-403.
32. Noordzij MJ, et al. Dermal factors influencing measurement of skin autofluorescence. Diabetes Technol Ther, 2011; 13(2): 165-70
33. Barski Oleg A, et al. Dietary Carnosine Prevents Early Atherosclerotic Lesion Formation in Apolipoprotein E–Null Mice. Arteriosclerosis, Thrombosis, and Vascular Biology, 2013; 33(6):1162-1170
34. Nagai K, et al. Role of L-carnosine in the control of blood glucose, blood pressure, thermogenesis, and lipolysis by autonomic nerves in rats: involvement of the circadian clock and histamine. Amino acids, 2012; 43(1):97-109
35. Aldini G, et al. The carbonyl scavenger carnosine ameliorates dyslipidaemia and renal function in Zucker obese rats. Journal of cellular and molecular medicine, 2011; 15(6): 1339-1354
36. Flancbaum L, et al. Existence of carcinine, a histamine-related compound, in mammalian tissues. Life Sciences, 1990; 47(17):1587-1593
37. Bellia F, et al. Neuroprotective features of carnosine in oxidative driven diseases. Mol Aspects Med, 2011; 32(4-6): 258-66
38. Gariballa SE, Sinclair AJ. Carnosine: physiological properties and therapeutic potential. Age Ageing, 2000; 29(3):207-10
39. Abe H. Interorgan transport and catabolism of carnosine and anserine in rainbow trout. Comparative Biochemistry and Physiology Part B: Comparative Biochemistry, 1991; 100(4): 717-720
40. Johnson P, et al. Regulation of muscle phosphorylase activity by carnosine and anserine. Biochemical and Biophysical Research Communications, 1982; 109(3):769-775
41. Ohtsuka C, Sato Y, Takahashi H. Homocarnosine levels in cerebrospinal fluid of patients with infantile spasms under ACTH therapy. Brain Dev, 1983; 5(5):464-8
42. Greene SM, et al. Enhanced carnosine (β-alanyl-L-histidine) breakdown and histamine metabolism following treatment with compound 4880. European Journal of Pharmacology, 1984; 99(1):79-84
43. Gille JJP, et al. Effect of antioxidants on hyperoxia-induced chromosomal breakage in Chinese hamster ovary cells: protection by carnosine. Mutagenesis, 1991; 6(4):313-318

44. Gercken G, Trotz M. Fatty acid synthesis in the arrested rabbit heart during ischemia. Pflügers Archiv, 1983; 398(1): 69-72
45. Boldyrev AA, et al. The antioxidative properties of carnosine, a natural histidine containing dipeptide. Biochemistry international, 1987; 15(6):1105-1113
46. Zhao J, et al. Carnosine protects cardiac myocytes against lipid peroxidation products. Amino Acids, 2019; 51(1):123-138
47. Yan SL, et al. Protective effects from carnosine and histidine on acetaminophen-induced liver injury. J Food Sci, 2009; 74(8):H259-65
48. Houjeghani S, et al. l-Carnosine supplementation attenuated fasting glucose, triglycerides, advanced glycation end products, and tumor necrosis factor-α levels in patients with type 2 diabetes: a double-blind placebo-controlled randomized clinical trial. Nutr Res, 2018; 49:96-106
49. de Courten B, et al. Effects of carnosine supplementation on glucose metabolism: Pilot clinical trial. Obesity (Silver Spring), 2016; 24(5):1027-34
50. Yılmaz Z, et al. The effect of carnosine on methylglyoxal-induced oxidative stress in rats. Arch Physiol Biochem, 2017; 123(3):192-198
51. Soliman KM, A.M.M.a.N.S.M. Attenuation of Some Metabolic Deteriorations Induced by Diabetes Mellitus Using Carnosine. Journal of Applied Sciences, 2007; 7:2252-2260
52. Lee YT, et al. Histidine and carnosine delay diabetic deterioration in mice and protect human low density lipoprotein against oxidation and glycation. Eur J Pharmacol, 2005; 513(1-2):145-50
53. Nagai K, et al. Possible Role of L-Carnosine in the Regulation of Blood Glucose through Controlling Autonomic Nerves. Experimental Biology and Medicine, 2003; 228(10):1138-1145
54. Menon K, Mousa A, de Courten B. Effects of supplementation with carnosine and other histidine-containing dipeptides on chronic disease risk factors and outcomes: protocol for a systematic review of randomised controlled trials. BMJ open, 2018; 8(3):e020623-e020623
55. Elbarbary NS, et al. The effect of 12 weeks carnosine supplementation on renal functional integrity and oxidative stress in pediatric patients with diabetic nephropathy: a randomized placebo-controlled trial. Pediatr Diabetes, 2018; 19(3):470-477
56. Cararo JH, et al. Carnosine and Related Peptides: Therapeutic Potential in Age-Related Disorders. Aging Dis, 2015; 6(5): 369-79
57. Sadowska-Bartosz I, Bartosz G. Prevention of protein glycation by natural compounds. Molecules, 2015; 20(2): 3309-34
58. Matsuzawa Y, Funahashi T, Nakamura T. The concept of metabolic syndrome: contribution of visceral fat accumulation and its molecular mechanism. J Atheroscler Thromb, 2011; 18(8):629-39
59. Miyatake N, et al. Reducing waist circumference by at least 3 cm is recommended for improving metabolic syndrome in obese Japanese men. Diabetes Res Clin Pract, 2008; 79(2): 191-5
60. Rothberg AE, et al. Impact of weight loss on waist circumference and the components of the metabolic syndrome. BMJ Open Diabetes Res Care, 2017; 5(1):e000341
61. Mong MC, Chao CY, Yin MC, Histidine and carnosine alleviated hepatic steatosis in mice consumed high saturated fat diet. Eur J Pharmacol, 2011; 653(1-3):82-8
62. Kawamoto R, et al. Relationships between lipid profiles and metabolic syndrome, insulin resistance and serum high molecular adiponectin in Japanese community-dwelling adults. Lipids Health Dis, 2011; 10:79
63. Ono T, et al. The Fasting-plasma Glucose Range in which Insulin Resistance Measured by Homeostasis Model Assessment Correlates with Euglycemic Clamping. 1999