

Glycophospholipid Formulation with NADH and CoQ10 Significantly Reduces Intractable Fatigue in Western Blot-Positive ‘Chronic Lyme Disease’ Patients: Preliminary Report

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Abstract

Background: An open label 8-week preliminary study was conducted in a small number of patients to determine if a combination oral supplement containing a mixture of phosphoglycolipids, coenzyme Q10 and microencapsulated NADH and other nutrients could affect fatigue levels in long-term, Western blot-positive, multi-symptom ‘chronic Lyme disease’ patients (also called ‘post-treatment Lyme disease’ or ‘post Lyme syndrome’) with intractable fatigue.

Methods: The subjects in this study were 6 males (mean age = 45.1 ± 12.4 years) and 10 females (mean age = 54.6 ± 7.4 years) with ‘chronic Lyme disease’ (determined by multiple symptoms and positive Western blot analysis) that had been symptomatic with chronic fatigue for an average of 12.7 ± 6.6 years. They had been seen by multiple physicians (13.3 ± 7.6) and had used many other remedies, supplements and drugs (14.4 ± 7.4) without fatigue relief. Fatigue was monitored at 0, 7, 30 and 60 days using a validated instrument, the Piper Fatigue Scale.

Results: Patients in this preliminary study responded to the combination test supplement, showing a 26% reduction in overall fatigue by the end of the 8-week trial ($p < 0.0003$). Analysis of subcategories of fatigue indicated that there were significant improvements in the ability to complete tasks and activities as well as significant improvements in mood and cognitive abilities. Regression analysis of the data indicated that reductions in fatigue were consistent and occurred with a high degree of confidence ($R^2 = 0.998$).

Conclusions: The combination supplement was a safe and effective method to significantly reduce intractable fatigue in long-term patients with Western blot-positive ‘chronic Lyme disease.’

Keywords: Lyme disease, Lipid Replacement Therapy, NT Factor, mitochondria, chronic fatigue, NADH, coenzyme Q10

BACKGROUND:

‘Chronic Lyme disease’ (sometimes called ‘post-Lyme syndrome’ or ‘post-treatment Lyme disease syndrome’) is a heterogeneous, chronic, systemic, multi-symptom illness resulting from the tick-borne infection of the spirochete *Borrelia burgdorferi* and associated co-infections (*Mycoplasma* species, *Bartonella* species, *Babesia* species, etc.) [1-3]. Although the early stages of this complex infection are widely recognized and routinely treated, there are controversies surrounding the diagnosis and treatment of advanced multi-symptom ‘post-Lyme disease syndrome,’ ‘post-treatment Lyme disease syndrome’ or ‘chronic Lyme disease’ [2-4]. One of the most common symptoms is chronic fatigue [2, 4, 5], and some patients with ‘chronic Lyme disease’ are initially diagnosed with another fatiguing illness, chronic fatigue syndrome (CFS) [6, 7]. Like ‘chronic Lyme disease,’ CFS is also characterized by multiple symptoms and chronic infections [7-10], and chronic fatigue is one of the most characteristic symptoms in both of these chronic illnesses [2, 4-7, 9].

INTRODUCTION:

Natural supplements have been used to reduce fatigue in patients with a variety of chronic illnesses [11, 12]. Few of these natural supplements were, however, considered effective [12]. Chronic or intractable fatigue that is not reversed by sleep is the most common complaint of patients seeking general medical care [13, 14]. Fatigue is quite common; it occurs naturally during aging and in many chronic conditions [14].

Fatigue has been described as a multidimensional sensation, and clinical studies have determined the extent of chronic fatigue in aging and in various medical conditions [13, 15-17]. Fatigue is perceived to be a loss of overall energy and inability to perform even simple tasks without exertion [13, 14]. At the cellular level fatigue is related to cellular energy systems found primarily in the cellular mitochondria and specifically in the inner mitochondrial membrane electron transport chain [18, 19]. Damage to mitochondrial membranes occurs mainly by oxidation, and this can result in increased ion leakage across mitochondrial membranes and impairment in the ability of mitochondria to produce high-energy molecules. Other cellular membranes are also affected by oxidation, resulting in impaired function [20, 21].

Damage to cellular membranes occurs mainly due to the production of free-radical oxidative molecules, such as reactive oxygen and nitrogen species (ROS/RNS). ROS/RNS include nitric oxide, oxygen and hydroxide radicals and other molecules that can cause oxidative stress and cellular damage, especially to sensitive membrane phospholipids, proteins and DNA [20-23]. Important targets of ROS/RNS damage are cellular membranes, and in particular, mitochondrial membranes, especially their phospholipid components [21-24]. In fatiguing

illnesses, such as CFS, patients show excess oxidative stress and increased susceptibility to peroxidation of membrane lipids [25, 26]. To some degree this excess oxidative stress can be reduced with antioxidant supplements [27-29], but these antioxidants cannot repair the oxidative damage done to cells. Thus supplements have been developed that contain unoxidized precursor molecules that can repair and prevent damage to cellular membranes.

Recent clinical trials have shown the effectiveness of lipid replacement therapy (LRT) plus antioxidants in the treatment of CFS and related conditions [16, 30]. LRT results in the actual replacement of damaged cellular lipids with undamaged (unoxidized) lipids to ensure proper function of cellular structures, mainly cellular and organelle membranes. LRT can result in the cellular delivery of unoxidized, undamaged membrane glycopospholipids in order to replace damaged lipids and restore function to oxidized cellular membranes.

Here we tested the results of using a formulation of glycopospholipids, CoQ10 plus microencapsulated NADH and other nutrients on suppression of fatigue during an open label two-month trial. The subjects in this study were ‘chronic Lyme disease’ patients defined as Lyme Western blot-positive patients with long-term intractable fatigue and other symptoms that had tried unsuccessfully many drugs and supplements to reduce their fatigue.

SUBJECTS AND METHODS:

Methods. An open label, institutionally approved clinical trial was initiated to study the effects of an all-natural supplement on fatigue in patients with Western blot-positive ‘chronic Lyme disease.’ The supplement product, ATP Fuel®, containing NT Factor®, microencapsulated NADH, CoQ10, pro- and pre-biotics and other nutrients (Researched Nutritionals, Inc., Los Olivos, CA), is a patent-pending proprietary nutrient complex containing an exogenous source of polyunsaturated phosphatidylcholine, phosphatidylglycerol, phosphatidylserine, phosphatidylinositol, and other membrane phospholipids, as well as coenzyme Q10 (CoQ10), microencapsulated reduced nicotinamide adenine dinucleotide (NADH) and other micronutrients (Table 1). The participants took the suggested daily dose (5 capsules in the morning and 5 at night) for 8 weeks.

Table 1. Test Supplement (ATP Fuel® with NT Factor®, NADH and CoQ10)^a

Component	Amount Per Serving^b	% Daily Value^c
Vitamin E (as alpha-D-tocopheryl)	20 IU	67
Krebs Energy Foundation TM NADH, Coenzyme Q10	35 mg	*
Mitochondria Pro Regulator TM	260 mg ^d	*

Calcium (as calcium pyruvate, calcium ascorbate dicalcium phosphate), phosphate/sulfate/pyruvate;
Phosphorus (as dicalcium phosphate);
Magnesium (as magnesium oxide)

Krebs Cycle Glucose Absorb TM Alpha-ketoglutaric acid, L-tyrosine	180 mg	*
RN Fatty Acid Metabolizer TM L-carnitine L-tartrate, pantethine	160 mg	*
NT Factor ^{®c} Maximum Potency TM	2,000 mg	*

^aATP Fuel[®] is a registered trademark of Researched Nutritionals, Inc., Los Olivos, CA.

^bOne Serving = 5 capsules; total protein per serving = 0.7 g. Total calories per serving = 9.5; 2.5 calories per serving from unsaturated lipids. Daily dose = 2 servings

^cDaily values are based on a 2,000 calorie per day diet.

^dPer serving: calcium 136 mg, iron 40 mcg, phosphorus 48 mg, magnesium 43 mg, manganese 10 mcg, potassium 9.5 mg

^eNT Factor[®] is a nutrient complex containing membrane polyunsaturated phospholipids extracted from soy lipids and purified using proprietary processes, Bifido and Lactobacillus bacteria with supportive essential growth media and phytonutrients for optimal absorption and performance, by Nutritional Therapeutics, Inc., Commack, NY.

*Daily values not established

Fatigue was scored using the validated Piper Fatigue Scale (PFS) [30-32]. The PFS is composed of 22 numerically scaled questions rated from 0 (no fatigue) to 10 (severe fatigue) [31, 32]. These items measure four dimensions of subjective fatigue: behavioral/severity (6 items); affective/meaning (5 items); sensory (5 items); and cognitive/mood (6 items). These are used to calculate the four sub-scale/dimensional scores and the total fatigue scores. The standardized alpha (Cronbach's alpha) coefficient of reliability did not drop below 0.99 for any of the subscale data, and the standard alpha for the entire scale of 22 questions was 0.98, indicating excellent reliability and internal consistency for this established instrument [33]. The study participants took the PFS survey at days 0, 7, 30 and 60.

Subjects. Participants were prescreened after an initial contact to determine whether they could have 'chronic Lyme disease' [34]. Subjects were included in the trial if they had been ill for at least 6 months, had a multi-symptom chronic condition characterized by several signs and symptoms, such as intractable fatigue, joint and muscle pain (arthralgia), muscle aches (myalgia), memory impairment and other symptoms [34], and they or their physician could provide Western blot evidence for Lyme disease *Borrelia burgdorferi* infection [2, 7, 34]. This was defined as at least three bands positive in the *Borrelia* Western blot. Those who qualified

completed an Informed Consent document and PFS on-line evaluation [30-32]. Participants were also asked how long their condition existed, whether it had been affected by any drug or supplement, and how many physicians they had been to before entering the trial. They were also asked if they were currently using any prescription medications to see if this might exclude them from the study, as determined previously [30, 35]. The completed online surveys were scored as described previously [35, 36].

There were 16 respondents (10 females and 6 males) who met the criteria of chronic Lyme disease and were fully compliant and completed the study. The mean age \pm SD of participants completing the study was 52.4 ± 10.8 (females, 54.6 ± 7.4 years and males, 45.1 ± 12.4 years, respectively).

Study Design. Each participant was given instructions to take the glycopospholipid supplement with NADH and CoQ10 daily after an initial PFS assessment (time 0). All subjects repeated the PFS assessment at the end of the first week on line without access to their previous scores. After the participants completed their PFS questionnaires at 30 and 60 days, all of the electronic forms were checked for verification, completion and scoring accuracy [36]. Compliance was determined by counting the remaining capsules left over by each participant in the study.

Statistics. Data were analyzed by ANOVA, with significance defined as $p < 0.05$. Further data analysis was performed with Tukey test and linear regression analysis, with significance defined as $p < 0.05$. The standardized alpha (Cronbach's alpha) was used to confirm reliability and internal consistency of the data [33].

RESULTS:

Subjects in the Study. The subjects in this study were *Borrelia* Western blot-positive, chronic disease patients who had moderate to severe chronic fatigue for years without improvement (mean \pm SD years without improvement of fatigue = 12.7 ± 6.6 years). Most had been to multiple physicians (mean \pm SD = 13.3 ± 7.6) and had taken a variety of remedies, drugs and supplements (mean \pm SD = 14.4 ± 7.4) without expected relief of their fatigue symptoms. Of the 20 subjects that started the trial, 16 completed the study. Almost all of the subjects that withdrew from the study did so without ever completing the day 0 PFS survey and taking the test supplement, or they were omitted from the data analysis for non-compliance during the trial.

Effects of the Test Supplement on Fatigue. We examined the effects of the combination glycopospholipid-NADH-CoQ10 preparation used in this study (Table 1) on fatigue scores in a 2-month open label trial and found significant improvements in the overall fatigue scores of the study subjects as measured by the PFS (Table 2). The initial PFS group mean total fatigue score \pm SEM was 8.1 ± 1.4 , and after 8 weeks of supplement this improved to 6.0 ± 1.2 , or a 26% reduction in fatigue. The mean decrease in fatigue scores was significant by t-test ($p < 0.0003$) and Wilcoxon signed-rank ($p < 0.0002$) analyses.

The PFS can be further dissected into subcategories that include: behavior/severity, affective meaning, sensory and cognitive/mood (Table 2). All of these subcategories showed

significant reductions at the end of the 8-week trial, indicating that there were significant improvements in all subcategories of fatigue. For example, there was a 26% reduction ($p < 0.005$) in severity/behavior of fatigue, indicating that there was a significant reduction in the intensity of fatigue, and a significant increase in the ability to complete tasks, socialize and engaging in sexual and other activities. Also, there was a 25% improvement ($p < 0.010$) in mood and cognitive ability, such as the ability to concentrate, remember and think clearly (Table 2).

Table 2. Overall Fatigue and Subcategories of the Piper Fatigue Scale Survey

Category	Mean Fatigue Level \pm S.E.M.		Percent Reduction	t-test
	Day 0	Day 60		
Overall Fatigue	8.1 \pm 1.4	6.0 \pm 1.2	26	p < 0.0003
Behavior/Severity ^a	8.5 \pm 1.7	6.3 \pm 1.2	26	p < 0.005
Affective/Meaning ^b	8.8 \pm 1.6	7.0 \pm 1.6	21	p < 0.010
Sensory ^c	8.4 \pm 1.2	6.2 \pm 1.3	26	p < 0.005
Cognitive/Mood ^d	7.0 \pm 1.4	5.3 \pm 1.2	25	p < 0.010

^aBehavior/Severity subcategory deals with completing tasks, socializing, engaging in sexual activity and other activities and intensity or degree of fatigue.

^bAffective/Meaning subcategory determines whether fatigue/tiredness is pleasant/unpleasant, patient is agreeable/disagreeable, protective/destructive, or feels normal/abnormal.

^cSensory subcategory determines whether patient is strong/weak, awake/sleepy, refreshed/tired, or energetic/unenergetic.

^dCognitive/Mood subcategory assesses whether patient feels relaxed/tense, exhilarated/depressed, able/unable to concentrate, remember and think clearly.

Regression Analysis of Data. To see if the trends in fatigue reduction over time during the trial were consistent, occurred with a high degree of confidence and could predict further reductions we conducted regression analyses of the data. As shown in Figure 1, the regression analysis of overall fatigue in each of the subcategories of fatigue (behavior/severity, affective meaning, sensory and cognitive/mood) indicated significant and consistent downward trends in the fatigue data ($R^2 = 0.998$), suggesting that further reductions in fatigue would have been likely if the trial had been continued (Figure 1). The regression R^2 values for the various subcategories were: Behavior/severity, 0.999; Affective meaning, 0.998; Sensory, 0.998; and Cognitive/mood, 0.999.

Regression analysis of the overall fatigue yielded $R^2 = 0.998$. This indicates a high level of confidence in the downward trends in all fatigue subcategories and total fatigue.

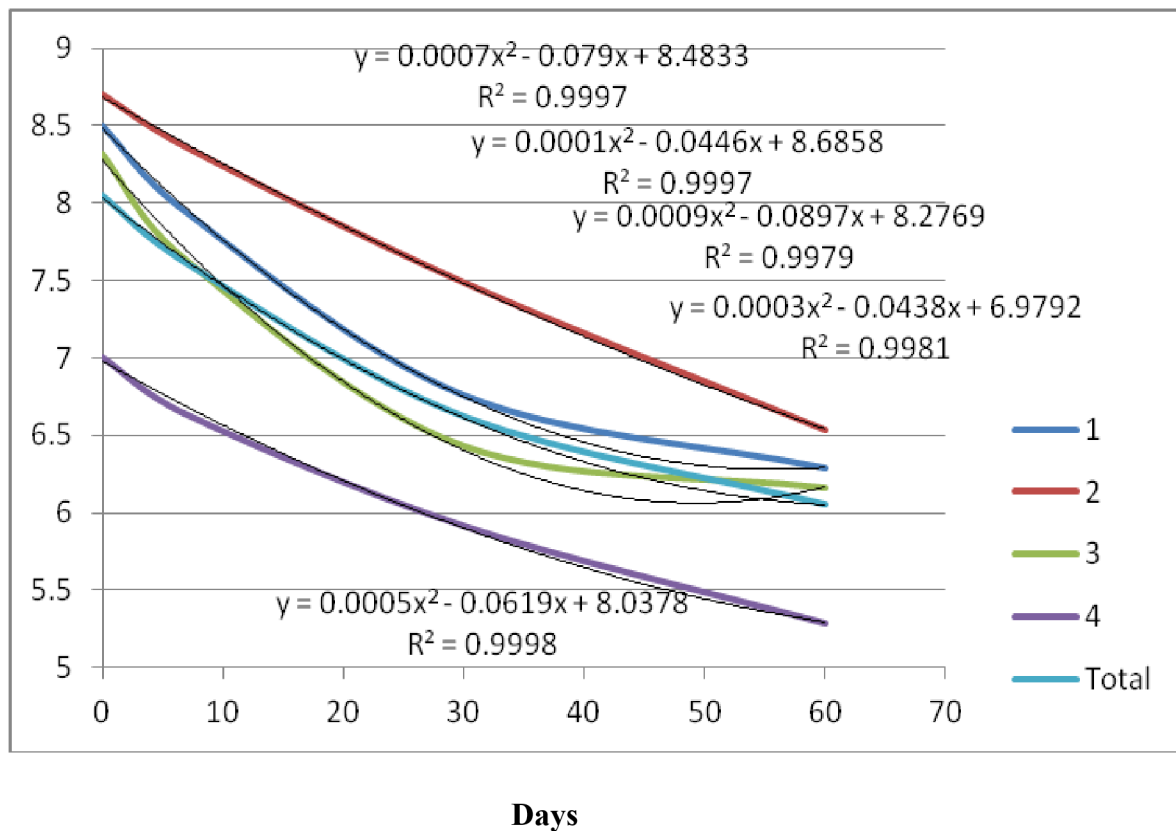


Figure 1. Data regression analyses of Lyme disease patients taking the test supplement in a 60-day trial. X axis, days; Y axis, Piper Fatigue Score. The regression R^2 values for the various subgroups were: 1: Behavior/severity, 0.999; 2: Affective meaning, 0.999; 3: Sensory, 0.998; and 4: Cognitive/mood, 0.999. Total: regression analysis of the overall fatigue, $R^2 = 0.998$.

DISCUSSION:

Lyme disease, the most common vector-borne infection in North America, is characterized by multiple signs and symptoms [1-3, 5, 34, 37]. Among the more commonly found symptoms are: joint and muscle pain (arthralgia), muscle aches (myalgia), memory impairment and fatigue [1, 5, 34]. These symptoms overlap significantly with CFS, which may be why some Lyme disease patients are initially diagnosed with CFS [6, 7]. Although the treatment of the early stages of Lyme disease are relatively straight forward and involve short courses of antibiotics [2, 3, 34, 37, 38], treatment of 'chronic Lyme disease' has proven to be particularly difficult, lengthy and controversial, involving complex regimens of antibiotics, herbal remedies, nutritional supplements and rehabilitation [34, 39]. Use of nutritional supplements has been recommended in the supportive treatment of 'chronic Lyme disease' [34, 39-41], but few if any clinical trials have examined the effectiveness of nutritional supplements on the signs and symptoms of Lyme disease [39].

Among the nutritional supplements suggested for the treatment of Lyme disease, omega-3 fatty acids, CoQ10, L-carnitine and NT Factor® have been suggested [39, 41]. NT Factor® is

of particular interest, since it has been used successfully in several animal and clinical studies [16, 22, 30, 35, 36, 43, 44]. In this formulation encapsulated lipids are protected from oxidation in the gut and can be absorbed and transported into tissues without oxidative damage. NT Factor® contains a variety of components, including phospholipids, glycopospholipids and other lipids, nutrients, probiotics, vitamins, minerals and other nutrients [22, 30, 35].

NT Factor® has been used in studies with severe chronic fatigued patients to reduce their fatigue [22, 30, 35, 43]. We found that fatigue was significantly reduced from severe to moderate fatigue, after eight weeks of supplementation with NT Factor® [30, 35, 43]. For example, use of NT Factor® for 12 weeks resulted in 35.5% reduction in fatigue ($p < 0.001$). In this clinical trial there were good correlations between reductions in fatigue and increases in mitochondrial function. After 8 weeks of LRT with NT Factor®, mitochondrial function was significantly improved ($p < 0.001$), and after 12 weeks of NT Factor® supplementation, mitochondrial function was found to be similar to that of young healthy adults [43]. After 12 weeks of supplement use, subjects were placed on placebo without their knowledge for an additional 12 weeks, and their fatigue and mitochondrial function were again measured. After the 12-week placebo cross-over period fatigue increased and mitochondrial function decreased, indicating that the NT Factor results on fatigue were not due to a “placebo effect” [43].

The dietary supplement we used was a preparation containing NT Factor®, CoQ10 and microencapsulated NADH. Microencapsulated NADH alone had been found to have a positive effect on fatigue in some patients. In one study a subset of patients responded or the response was for a limited period of time. In a randomized, cross-over study on CFS patients 8 of 26 (30.7%) responded to microencapsulated NADH compared to 2 of 26 (8%) on placebo ($p < 0.05$) [45]. These results were not considered significant by others [46, 47]. In a 24-month trial Santaella et al. [35] compared the effectiveness of oral NADH to combination psychological/nutritional therapy in 31 CFS patients. They found that NADH resulted in a reduction in fatigue but only in the first trimester of the trial. In subsequent trimesters, symptom scores were similar in the NADH and psychological/nutritional arms of the trial. In another study oral NADH was given for two months to CFS patients. In this study the NADH group showed a decrease in anxiety and maximum heart rate after a stress test compared to the placebo group, but differences were not found in the functional impact of fatigue, quality of life, sleep quality, exercise capacity or functional reserve [49].

NADH is required as an important cofactor in many cellular enzyme pathways, including those involving certain transferases, polymerases, synthases, deacetylases and other enzymes [50]. It is also important in signal transduction, DNA repair, salvage pathways and post-translational protein modifications [51]. NADH in its reduced form is an important component of the mitochondrial electron transport chain, and it plays an important role in programmed cell death [52, 53].

In the mitochondrial electron transport chain CoQ10 acts as an essential electron carrier, and it is also an important antioxidant as well as a molecule involved in gene regulation [54, 55]. It has been used as an important supplement in a variety of chronic illnesses and age-related conditions [55-57]. In our studies CoQ10 was used to improve energy transduction and combat oxidative stress [55, 58], while CoQ10 has been used extensively for combined nutraceutical therapy in mitochondrial cytopathies [58, 59].

Here we used an all-natural formulation containing NT Factor®, CoQ10, NADH and other nutrients in ‘chronic Lyme disease’ patients who had intractable chronic fatigue for an average of over 12 years. Chronic fatigue is known to cause loss of mitochondrial function [59, 60]. These patients had tried a large number of supplements and drugs (14.4 ± 7.4) with little or no effect on their fatigue. Although these patients had intractable fatigue, they responded well to the combination supplement containing NT Factor®, CoQ10 and NADH. Regression analysis indicated that the data were consistent and reliable with a high degree of confidence ($R^2 = 0.998$). The regression analysis further suggested that continuing the supplement beyond the 60 days of the trial could result in further reductions in fatigue.

Finally, the supplement was effective and safe for ‘chronic Lyme disease’ patients. This preliminary study should be repeated with a longer test period, a control arm and larger numbers of fatigued patients with other chronic illness diagnoses. Although there is continuing controversy surrounding the evidence for chronic *Borrelia* infection in ‘chronic Lyme disease’ or ‘post-treatment Lyme disease,’ and the treatment of potential chronic infections [2-5, 34, 37, 61], there is general agreement that these patients have chronic fatigue symptoms that should be addressed in any treatment strategy [34, 37, 41]. This preliminary study was only directed at reducing fatigue and cannot address other controversies surrounding ‘chronic Lyme disease’ or ‘post-treatment Lyme disease’ treatments [3, 37, 61].

CONCLUSIONS:

After 60 days use of a supplement containing a mixture of phosphoglycolipids, CoQ10 and NADH, fatigue measured with the validated Piper Fatigue Scale was significantly reduced in patients with intractable fatigue and Western blot-positive ‘chronic Lyme disease.’ The supplement was safe and effective and for use in these patients, but the study should be repeated with larger numbers of patients for a longer period of time.

Competing Interests:

The authors have no financial interests or conflicts of interest.

Authors’ Contributions:

All authors contributed to this study.

Abbreviations:

CoQ10, coenzyme Q10; CFS, chronic fatigue syndrome; LRT, Lipid Replacement Therapy; NADH, reduced nicotinamide adenine dinucleotide; PFS, Piper Fatigue Scale; ROS/RNS, reactive oxygen and nitrogen species

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