L-Carnitine in Acute Phosphide Pesticide Poisoning: A Randomized, Clinical Trial

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Abstract: Metallic phosphides are extremely toxic pesticides & phosphides poisoning is commonly encountered in emergency departments in most developing countries. Research interest in L-carnitine has grown among increasing evidence of the role of oxidative stress in phosphides poisoning. We aimed to assess the efficacy and safety of L-carnitineas an adjuvant treatment in patients with acute phosphide poisoning. This was a randomized, controlled, parallel-group trial on 50 patients suffering from acute phosphide poisoning, who were admitted to the Poison Control Center (Emergency Hospitals, Tanta & Mansoura Universities), Tanta and Mansoura, Egypt, between January 2016 to January 2018. Interventions included intravenous L-carnitine (1 g/ 8 hours) as an added treatment to the conventional measures versus only the conventional treatment. Outcome measures included mortality, duration of hospitalization and the need for mechanical ventilation. No significant difference was found between both groups regarding demographic characteristics, toxicological and clinical data as well as routine laboratory investigations. No adverse effects to L-carnitine therapy were reported. Malondialdehyde significantly decreased and reduced glutathione & total antioxidant capacity significantly increased only in the LC-treated patients. The length of hospital stay showed significant difference between both groups. From this study, it could be concluded that LC had a good antioxidant effect and can safely be used as a promising adjuvant therapy in the treatment of acute phosphide poisoning.

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1. Introduction

Metallic phosphides are extremely toxic pesticides that are regulated in their usage [1]. Aluminum and zinc phosphides are highly effective insecticides and rodenticides which are used widely to protect grain in stores and during its transportation. Phosphides poisoning is commonly encountered in emergency departments in most developing countries [2].

Aluminum phosphide has currently generated interest with increasing number of cases in the past four decades because of its increased use for agricultural and nonagricultural purposes. Its easy availability in the markets has led to its increased misuse to commit suicide. Ingestion is usually suicidal, uncommonly accidental and rarely homicidal [3].

In Egypt in **1993 Abdelmagid and Salem** [4] surveyed 5913 patients admitted to the Alexandria Poison Center during one year and recorded that pesticide poisoning accounted for 14.3% of admitted cases. In 2006, at Ain Shams University poisoning center, insecticide poisoning represented 51.0% of admitted cases (a total of 21,805 cases); of this number, organophosphorus insecticides accounted for 75.0%, zinc phosphide 20.0% and carbamates 5.0% [5]. In 2018, at Menoufia Poison & Dependence Control Center (MPDCC), insecticides poisoning represented 15.5% of admitted cases (a total of 2026 cases); of this number, phosphides poisoning accounted for 5.6% [6].

Various mechanisms have been described for phosphides toxicity. Cellular damage and cardio-respiratory failure are the most common mechanisms of mortality and morbidity after poisoning [7]. Phosphine acts as a strong reducing agent and noncompetitively inhibits mitochondrial respiratory chain enzyme cytochrome C oxidase leading to the generation of reactive oxygen species and cellular peroxides [8]. Oxidative degradation of lipids known as lipid peroxidation, and other oxidant mechanisms damage biological macromolecules specially the cell membrane ultimately leading to cell death [9].

L-carnitine (LC) is a non-protein amino acid that is synthesized from the essential amino acids lysine and methionine. Many *in vitro* and animal studies have reported that LC is a free radical scavenger, which protects antioxidant enzymes from oxidative damage.

In a human study in 2011, Cao et al. administered LC supplement to healthy volunteers and observed that LC significantly increased the levels of antioxidant enzymes activities [10].

Therefore, LC, being a widely available, inexpensive drug with a prominent antioxidant activity, was evaluated as an adjuvant therapy in treatment of patients with acute phosphide poisoning.

2. Patients and Methods

Patients: This randomized clinical trial was conducted on 50 patients suffering from acute phosphide poisoning admitted to the Poison Control Center (Emergency Hospital, Tanta & Mansoura University). from January 2016 to January. 2018 The study was approved from the Research Ethics Committee of Faculty of Medicine, Tanta University. An informed written consent was obtained from each patient or his/her guardian (if the patient was unable to participate in consent process) after receiving detailed information about the study. Confidentiality of the data was maintained by making code number for each patient (Research ethics committee approval number: 15/12/30628, Trial ID on Clinical Trials.gov PRS: NCT03953248).

Eligibility criteria: Patients (male or female, aged 12 years or older) with symptomatic acute phosphide poisoning (deliberate or accidental). Diagnosis was made on basis of: (i) Typical clinical manifestations due to and shortly after single exposure to phosphide. (ii) Reliable identification of phosphide based on container brought by patient attendants or a subsequent confirmation by silver nitrate test for phosphine detection in stomach contents [11]. Pregnant and lactating women, patients with ingestion or exposure to other substances in addition to the phosphide and those presenting more than 6 hours of exposure to the phosphide compound were excluded from this study. Neither analysis of the content of the containers brought nor measurement of the phosphide or its metabolites in the patient urine was performed.

Methods: The study volunteers (50 patients) were randomly allocated into two equal groups I (conventional treatment) and II (conventional treatment plus LC) using the sequentially numbered, opaque sealed envelopes method [12]. The envelopes were impermeable to intense light, and the allocation sequence was concealed from the physician enrolling and assessing participants. To prevent subversion of the allocation sequence, the name and hospital admission number of the participant were written on the envelope. Carbon paper transferred the information onto the allocation card inside the envelope. Corresponding envelopes were opened only after the enrolled participants completed all baseline assessments, and it was time to allocate the intervention. The study participants, health care providers and data analysis were kept blinded to the allocation.

All patients received the conventional treatment, which included all or some of the following as indicated: patient resuscitation, decontamination, adequate hydration and supportive treatment. In addition to the conventional treatment, group II received L-carnitine (L – Carnitine ampoule is produced by medical union pharmaceuticals for MEPACO – MEDIFOOD. Each package contains 5 ampoules (5ml). Each 5ml ampoule contains 1gm L – carnitine) intravenously, in a dose of 1 gm / 8 hours. This dose of LC has been reported to reduce oxidative stress and to improve antioxidant status. Patients were monitored with a detailed documentation of any adverse effect due to drug therapy.

All patients were subjected to full history taking (including age, gender, occupation, level of education, circumstances of poisoning, route of exposure, time interval between exposure and beginning of treatment and history of medical diseases) and complete physical examination (including level of consciousness by Glasgow coma scale, regular monitoring of vital signs and general clinical examination). Arterial blood samples were obtained from each patient for blood gas analysis, whereas venous samples were used for estimation of the biochemical profile, oxidative stress biomarkers including malondialdehyde (MDA) [13] total antioxidant capacity (TAC) [14] and reduced glutathione (GSH) [15].

All the patients were prospectively monitored by qualified physicians with regular measurement of their vitals and oxygen saturation via a bedside monitor, and they were followed up until discharge from the hospital. Adverse reactions to LC were recorded.

Outcome measures: This study was a clinical trial to reveal the safety and efficacy of LC as an added treatment in acute phosphide poisoning. The primary outcome was mortality, whereas secondary outcome measures included the duration of hospital stay, and the need for intubation.

Statistics: The data were analysed according to intention to treat approach involving all patients who were randomly assigned. The primary objective of this study was to compare the incidence of mortality (Yes/No) in the study treatment groups. Secondary continuous outcome, as length of hospital stay was analysed using the Mann–Whitney test. Baseline data of study participants were collected and summarized in the form of mean ± S.D. Also, the data were analysed using the t-test or the Mann–Whitney U-test based on normal distribution assumption for continuous data and Fisher's exact test for categorical variables to check for imbalance between treatment arms. The p-values of 0.05 or less were considered to

be statistically significant. Statistical analysis was carried out using Statistical Package for the Social Sciences (SPSS) for Windows, version 15.0 (SPSS, Chicago, IL, USA).

3. Results

Table 1 shows baseline demographic and clinical characteristics of the study groups. The groups were homogeneous regarding age and gender. No patient complained of coma. No significant difference was found between both groups regarding the clinical manifestations. No major adverse effects to LC were reported. There was no significant difference between the two studied groups regarding vomiting and abdominal pain.

Statistically significant differences were noticed between both groups in serum levels of each of the oxidative stress biomarkers MDA, TAC and GSH. Administration of LC therapy to patients of group II was associated with a significant reduction of MDA but a significant increase in TAC and GSH when the mean level of each biomarker was compared before and after LC therapy (table 2).

There was no significant difference between the two studied groups regarding survivaland need for intubation. Length of hospital stay showed significant difference between both groups (table 3).

Table 1. Baseline demographic and clinical characteristics.

	Group I (n=25) Standard treatment	Group II (n=25) LC + Standard treatment
Age (years)	26.6	25.3
Gender (female)	14	14
Suicidal	25	25
Delay time (hrs)	2	2
Vomiting	10 (40%)	11 (44%)
Abdominal pain	5 (20%)	4 (21%)
Pulse (bpm)	97.2	96
Systolic BP	89.5	96
Diastolic BP	57.5	56.7

Data are medians (interquartile range), means (S.D.) or numbers (%).

Table 2. Serum malondialdehyde (MDA), total antioxidant capacity (TAC) and blood reduced glutathione (GSH) levels.

	Groups	On admission (mean ± S.D.)	After treatment (mean ± S.D.)	p-Value
MDA (nmol/ml)	Group I (n=25)	14.8 ± 3.4	15.1 ± 5.8	0.589
	Group II (n=25)	15.3 ± 3.6	7.9 ± 2	<0.001*
TAC (mM/L)	Group I (n=25)	5.6 ± 1.5	6.3 ± 6.1	0.179
	Group II (n=25)	5 ± 1.6	10.2 ± 2.3	<0.001*
GSH (mmol/ml)	Group I (n=25)	2.5 ± 1	2.7 ± 0.9	0.436
	Group II (n=25)	2.3 ± 0.8	4 ± 0.8	<0.001*

Data are means (S.D.).

Table 3. Outcome measures.

	Group I (n=25) Standard treatment	Group II (n=25) LC+ Standard treatment	p-Value
Improved	5 (20%)	9 (36%)	0.208
Death	20 (80%)	16 (64%)	
Intubation	10 (40)	6 (24%)	0.225
Hospital stay (hrs)	7	18	0.023*

Data are medians (interquartile range), means (S.D.) or numbers (%).

4. Discussion

In this randomized trial, the baseline characteristics showed no significant difference between the study groups. No adverse effects to LC therapy were reported. Malondialdehyde significantly decreased and total antioxidant capacity & reduced glutathione significantly increased only in the LC-treated patients. no significant difference was recorded between the two studied groups regarding mortality and need for intubation. The length of hospital stay showed significant difference between both groups. Regarding vomiting and abdominal pain, the study did not find any significant difference between patients who received LC and those who did not receive LC therapy.

No significant difference could be detected between group I and group II patients regarding oxidative stress markers; malondialdehyde (MDA), total antioxidant capacity (TAC) and reduced glutathione (GSH) on admission. The results of oxidative stress markers obtained on admission in this study were comparable to results gathered from different studies [16] [17] [18] [19] [20]. Phosphide-induced oxidative stress had been established in rats [21], nematodes [22], mammalian cell lines [23] and in insects [24]. Most previous reports were confined to animal studies and experimental models [25].

In the current study, treatment with L - carnitine significantly decreased MDA and elevated TAC and GSH in group II compared to group I that received only routine treatment. Such change in oxidative stress parameters could be explained by antioxidative role of L-carnitine which acts as direct scavenger effect of reactive oxygen species [26].

Reduction in MDA level after L - carnitine treatment in the current study is in accordance with previous research works assessing different antioxidant modalities [27] [28]. Results from these modalities signified a role for N-acetyl cysteine (NAC), endogenous glutathione, melatonin and magnesium in phosphide-induced oxidative stress. These studies were experimental and few studies were on human models such as by **Chugh and his colleagues (1997b)** [29]**.** The other study was conducted in Poison Control Center (Emergency Hospital, Tanta University) by **El-Ebiary and Abuelfadl (2017)** [30]**.** They assessed the antioxidant effect of intravenous magnesium and NAC respectively in management of acute phosphide poisoning.

Significant changes in serum TAC was proved to be related to L-carnitine [31]. **Lee et al. (2014)** [10] reported that LC consumption resulted in significant rise in antioxidant enzymes activities. **Fatouros et al. (2010)** [32] found that LC intake could be beneficial in improving antioxidant status. Increase in TAC levels after supplementation with LC could be associated with suppression of xanthine oxidase system [33]. LC supplementation enhances antioxidants system components, such as glutathione peroxidase, vitamin A, E and C [34] [35] [31].

Ates et al. (2008) [36] found that LC treatment enhances plasma GSH level. This antioxidant effect may be due to the decrease of lipid peroxidation caused by L-carnitine treatment. **Yapar et al. (2007)** [37] reported that LC significantly reduces MDA concentrations and increases GSH levels.

It is expected to find a statistically significant increase in hospitalization time in group II compared to group I. This might be because 36% of group II patients have survived, subsequently, have stayed in the hospital to complete their treatment. Meanwhile, 80% of group I patients have not survived and, subsequently, have not stayed in the hospital.

Conclusion

L-Carnitine may safely be used as an adjuvant to conventional treatment of acute phosphide poisoning as no major adverse effects were reported with its use. Further studies in a larger number of patients are required before a conclusion can be made about the efficacy of LC.

Limitations

The current study limitations include the small sample size; however, this was a pilot study assessing the safety and effectiveness of LC in acute phosphide poisoning, and the results of which should pave the way for larger trials recruiting patients based on sample size calculation.

Potential Conflict of Interest

The authors declare that they have no conflict of interests.

Financial Support

This study received no specific funding.

Ethical Approval

All procedures performed in this study were in accordance with the ethical standards of the institutional research committee and with the 1964 Helsinki Declaration and its later amendments.

Trial Registration

This trial is registered at the ClinicalTrials.gov PRS: NCT03953248.

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