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N-acetylcysteine Use in Treatment of Acute Aluminium Phosphide Poisoning: Systematic Review and Meta-Analysis

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Abstract

Background: Aluminum phosphide (AlP) is a popular used rodenticide. It inhibits oxidative phosphorylation, and causes depletion of glutathione, resulting in cellular wall dysfunction. N-acetylcysteine (NAC) is a glutathione precursor that would be effective in treatment of AlP poisoning. **Aim of the work:** Provide evidence based systematic review about role of NAC in treatment of AlP poisoning which may help in developing clear guidelines for treatment of such lethal poisoning. **Methodology:** We followed PRISMA guidelines during preparation of this study. PubMed, EKB, ScienceDirect and Cochrane CENTRAL were searched to identify the published literature from inception to June 2022. In addition, we searched for ongoing studies, reference lists for additional studies. We included randomized cotrolled trials (RCTs) and observational studies (OSs) published in English, those fulfilling inclusion criteria. **Results :** The study included four RCTs and two OSs with total 286 participants. The current study revealed that there was a significant reduction in mortality rate (OR 0.38, 95% CI [0,23 to 0.66]) and duration of hospital stay in survivors (SMD -1.73 days, 95% CI [-2.35, -01.11]) as well as a significant increase in survival time in non survivors in patients who received NAC, compared with those who did not receive NAC (SMD 0.87 day, 95% CI [0.37, 1.37]). There was no significant difference between NAC and control groups regarding the need for mechanical ventilation (OR 0.51, 95% CI [0,23 to 1.10]). **Conclusion:** N-acetylcysteine in treatment of acute AlP poisoning can reduce the mortality rate and duration of hospital stay in survivors and increase survival time.

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Key words Aluminium phosphide; Meta-analysis; N-acetylcysteine; poisoning, randomized controlled trials; Systematic review

Introduction

Rodenticides are considered a global challenge to public health. Annually, 250,000 to 370,000 people die from deliberate ingestion of pesticides, which is responsible for about one-third of suicidal attempts worldwide (Manouchehri, et al., 2019).

Phosphides are normally found as powders or pellets, usually in the form of zinc or aluminium phosphide (Zn3P2 and AlP, respectively), Calcium and magnesium phosphides are also available (Altintop and Tatli, 2017).

Aluminum phosphide is a highly popular indoor and outdoor pesticide used in many developing countries to protect grain in stores and during transportation. Even 500 mg of this compound can be fatal for humans with mortality rates as high as 70–100% in various studies (Nourbakhsh, et al., 2019).

The toxicity of aluminium phosphides is due to production of deadly phosphine gas in contact with water or diluted acids. Phosphine gas is typically produced within 30 minutes of phosphide consumption (Yan et al., 2018). The main mechanisms of toxicity are electron transfer blockage and non-competitive inhibition of cytochrome oxidase c, which inhibits oxidative phosphorylation, and in turn, cellular respiration resulting in activation of peroxide radicals. In addition, phosphine can inhibit catalase and deplete glutathione, resulting in cellular wall dysfunction (Ari et al., 2022).

Metal phosphides can result in serious systemic poisoning; cardiovascular collapse and cardiogenic shock may occur due to their direct effects on myocytes, intravascular fluid leakage into the third space, severe metabolic acidosis, and poor tissue perfusion (Bansal et al., 2017).

N-acetyl cysteine (NAC) is a novel thiol compound, commonly used as a mucolytic agent, and a precursor of L- cysteine and reduced glutathione (GSH). In addition, NAC is a source of sulfhydryl groups in cells and free radical scavenger as it interacts with reactive oxygen species (ROS) such as OH and H2O2 (Colovic et al., 2018).

Although NAC is widely known as an antidote to acetaminophen overdose, it has multiple other uses supported by various levels of evidence. These diverse clinical applications are linked to its ability to support the body's antioxidant and nitric oxide systems during

stress, infections, toxic assault, and inflammatory conditions (Tenório et al., 2021).

Although phosphide is well known as a lethal poison with neither an available effective antidote nor a specific treatment (Abdelhamid et al., 2023), in animal studies, NAC has been shown to have a protective role against phosphide-induced cardiovascular complications by protecting myocytes from the oxidative stress induced by phosphine, thus stabilizing blood pressure and pulse with dramatic improvement of outcome (Asghari et al., 2017). In addition, human studies revealed that NAC decreases mortality rates, length of hospitalization, and the frequency of intubation and mechanical ventilation after phosphide poisoning (ELabdeen et al., 2020).

So, it is important to do systematic review of the existing studies about NAC usage in acute aluminium phosphide poisoning to assess its efficacy in treating such lethal condition.

Aim of the Work

Provide evidence based systematic review about role of NAC in treatment of phosphide poisoning which may help in developing clear guidelines for treatment of such lethal poison.

Methodology

- Study design:

This is a systematic review and meta-analysis study. We followed PRISMA statement guidelines during preparation of this systematic review and meta-analysis.

- Criteria for considering studies for this review:
  - A. Inclusion criteria:
    1. Types of studies: We included all randomized controlled trials (RCTs) comparing acute AlP poisoning outcomes between groups received NAC or those did not. Since we expected to find very few of these, so we looked at observational studies, such as cohort studies, case-control, and cross-sectional studies.
    2. Types of participants: All acutely intoxicated patients with aluminium phosphide in the conducted studies regardless age and sex.
    3. Type of intervention: Use of NAC in hospitalized patients diagnosed as acutely intoxicated with aluminium phosphide.
    4. Types of outcome measures: We included studies reporting at least one of the following outcomes:
       - ⇒ Primary outcomes: Mortality and morbidity rates including cardiotoxicity, hepatotoxicity, and others.
       - ⇒ Secondary outcomes: Duration of hospitalization in survivors, duration of hospitalization in non survivors (survival time), and need for mechanical ventilation.
  - B. Exclusion criteria: Patients with history of cardiac, renal and hepatic diseases, opinion studies, studies conducted on animals, and studies not listed in inclusion criteria.

We included studies published or translated to English with no limits to age, sex, and publication time.

- Methods

I. Search methods for identification of studies

a) Electronic searches: We searched PubMed (from 1947 to 19 June 2022), Egyptian Knowledge Bank (EKB) (from 1918 to 19 June 2022), ScienceDirect (from 1989 to 19 June 2022) and CENTRAL (Cochrane Central Register of Controlled Trials) (from 2013 to 19 June 2022). We used a combination of the following keywords:

("Aluminium phosphide" OR phostoxin OR phosphine OR "rice tablet" OR rodenticide*) AND (N-acetylcysteine OR NAC OR antioxidant* OR "supportive measure*")

We followed the search tips of each database, and our searches were not restricted by language of publication.

b) Other search resources: We searched for ongoing clinical trials and unpublished trials via ClinicalTrials.gov (www.ClinicalTrials.gov) and the World Health Organization International Clinical Trials Registry Platform (www.who.int/ictrp). We also manually searched reference lists from the relevant articles of included studies, asking experts about additional studies, and attending conferences.

II. Data collection and analysis

a) Selection of studies: We merged search results using Endnote reference management software (Endnote 20) and removed duplicate records of the same report then examination of titles and abstracts using RAYYAN online application was done to remove obviously irrelevant reports (https://www.rayyan.ai). Moreover, full text examination of the potentially relevant reports was done by the author and the four supervisors for compliance of studies with eligibility criteria. Only articles fulfilling the inclusion criteria were included for further steps of data collection, analysis, and reporting. We recorded the selection process in detail to complete a PRISMA flow diagram.

b) Data extraction and management: Data were extracted independently by the first and the fifth authors and any discrepancies were resolved by five-member discussion and consultation with the original study. For missing information, we contacted the trial's authors for incomplete data.

We extracted the following study characteristics and outcome data from the included studies:

Methods; study design, study setting, date and duration of study, participants; mean age, age range, gender, severity of the condition, inclusion and exclusion criteria, intervention;

intervention, comparison, and any co interventions, outcomes; specified and collected outcomes, time points reported, notes; comments on quality of studies, notable conflicts of interest of trial authors, funding of trial.

III. Assessment of risk of bias in included studies: Risk of bias was assessed by the first author then revised by the other four authors according to recommendations of the Cochrane Handbook for Systematic Reviews of interventions *(Higgins et al., 2019).*
   A. Assessment of risk of bias in randomized controlled trials: We used COCHRANE ROB tool for randomized clinical trials studies. For each domain, we judged the risk of bias as low, high, or unclear if there was insufficient information to assess risk of bias. We resolved any disagreement with five-member discussion. The following definitions were used in the assessment of risk of bias in RCTs: random sequence generation (selection bias), allocation concealment (selection bias), blinding of participants and personnel (performance bias), blinding of outcome assessment (detection bias), incomplete outcome data (attrition bias), selective reporting (reporting bias), and other bias.

      If the trial had been assessed at low risk of bias in all the above domains, we judged it as having low risk of bias. If the trial had been assessed at unclear or high risk of bias in one or more of the above domains, we judged it as having high risk of bias.
   B. Assessment of risk of bias in observational studies: We also used Newcastle Ottawa Scale (NOS) to assess quality and risk of bias of observational studies. Newcastle Ottawa Scale is a 9-star scale for observational studies assessing the quality of selection (maximum 4 stars), comparability (maximum 2 stars), and outcome in cohort studies or exposure in case control studies (maximum 3 stars). We judged the study as good quality, fair quality or poor quality as follows: Good quality: 3 or 4 stars in selection domain AND 1 or 2 stars in comparability domain AND 2 or 3 stars in outcome/exposure domain; Fair quality: 2 stars in selection domain AND 1or 2 stars in comparability domain AND 2 or 3 stars in outcome/exposure domain; Poor quality: 0 or 1 star in selection domain OR 0 stars in comparability domain OR 0 or 1 star in outcome/exposure domain.

IV. Measures of outcomes: For evaluation of the dichotomous outcomes (mortality rate and need for mechanical ventilation), we recorded the total number of people with one or more events within each study and we presented comparisons between groups as odds ratio [with corresponding 95% confidence intervals (CIs)] instead of risk ratio to resolve heterogeneity that appeared when using risk ratio in meta-analysis. For continuous outcomes (duration of hospital stay in survivors and survival time in non survivors), we recorded the mean, standard deviation, and total number of people in both groups of each study, and we presented comparison between groups as standard mean difference [with corresponding 95% confidence intervals (CIs)].

V. Dealing with missing data: We contacted trial's authors for clarification about missing data in identified publication reports and then incorporated data when provided by the authors. We do all analyses according to the intention-to-treat principle by including all participants who were randomized in the statistical analysis and analyzing them according to the group they were originally assigned, irrespective of compliance or follow up *(McCoy, 2017).*

VI. Assessment of statistical heterogeneity: Heterogeneity which is a significant variation in the effect size of the included studies was assessed by the following tests: Cochrane Q chi square test: P-value < 0.1 is a statistically significant test, donated heterogeneity among the studies, and I-squared (I^2) index which is calculated as follows: $I^2 = \left(\frac{Q - df}{Q}\right) \times 100\%$ as Q is cochrane Q chi square and df is degree of freedom. The I-squared is interpreted as follows: 0% to 40%: might not be important. 30% to 60%: may represent moderate heterogeneity. 50% to 90%: may represent substantial heterogeneity. 75% to 100%: considerable heterogeneity *(Borenstein et al., 2019; Mohan and Adler, 2019).*

VII. Data synthesis
   A. Meta-analysis: We used review manager version 5.4 (RevMan 5.4) for data analysis. Dichotomous data was pooled as odds ratio (ORs) using the Mantel–Haenszel method and continuous data was pooled as standard mean difference (SMD) [with the respective 95% confidence intervals (CIs)] using Inverse Variance method. The analysis was conducted under the fixed-effect model. Forest plots were generated to illustrate the study-specific and pooled effect size. *P* value <0.05 was considered as statistically significant.
   B. Subgroup analysis: Subgroup analysis included the following: studies with different dosage forms and according to types of studies (randomized controlled trials versus observational studies).
   C. Publication bias: Publication bias assessment is not reliable for <10 pooled studies according to Egger and colleagues. Therefore, in the present study, we could

not assess the existence of publication bias by Egger's test for funnel plot asymmetry *(Egger et al., 1997).*

- Summary of finding and assessment of the certainty of the evidence: We assessed confidence in the evidence from included RCTs using GRADE criteria (GRADEpro GDT) an online guideline development tool (http://gradepro.org), and we constructed 'Summary of findings' table that included our review outcomes and comparisons. We assessed five factors referring to limitations in the study design and implementation of included studies that suggest a high likelihood of bias: Study risk of bias, indirectness of evidence (population, intervention, control, outcome), unexplained heterogeneity or inconsistency of results, imprecision of results (wide confidence intervals), and high probability of publication bias *(Schünemann et al., 2020)*.

The certainty of evidence is defined as the following: High certainty, we are very confident that the true effect lies close to that of the estimate of effect. Moderate certainty, we are moderately confident of the effect estimate. Low certainty, our confidence in the effect estimate is limited. Very low certainty, we have very little confidence in the effect estimate.

Results

We included six studies in our review; four randomized controlled trials *(Tehrani et al., 2013; Bhalla et al., 2017; El-ebiary and Abufad, 2017; Emam et al., 2020)* and two observational studies; a cohort study *(Agrawal et al., 2014),* and a case-control study *(Taghaddosonijad et al., 2016)* with total 286 participants of whom 145 received NAC. Four of the included studies *(Agrawal et al., 2014; Taghaddosonijad et al., 2016; Bhalla et al., 2017; Emam et al., 2020)* used NAC with a dose of 300 mg/kg intravenous over about 20 to 21 hour (continous infusion or divided as150 mg/kg over one hour then 50 mg/kg over four hours then 100 mg/kg over 16 hours) and the two remainder studies *(Tehrani et al., 2013; El-ebiary and Abufad, 2017)* used intravenous NAC with a dose of 1.33gm/kg over 72 hour divided as (140 mg/kg as a loading dose then 70 mg/kg every four hours for 17 doses). The included studies were published between 2013 and 2020. Two studies were conducted in India, two in Iran, and two studies were carried out in Egypt. Baseline characteristics of the populations of the included studies are shown in (Table 1) and the summary of their designs and their main results are shown in (Table 2).

Three studies were excluded with reasons as follows; *Bhat and Kenchetty (2015)* was about rodenticides in general and AlP was not specified; *Abdel-hady et al., (2019)* had no details about the participants who received NAC, no additional data were received when contacted trials' authors; *Tawfik (2020)* was an abstract, so we contacted the author who send us the original study which was about metal phosphide poisoning (aluminium phosphide and zinc phosphide) and there were no isolated data about AlP alone. In addition, three ongoing studies (Irct20200724048192N1, NCT04509258, and NCT05370729) were excluded.

I. Results of the search

The results of our searches are detailed in a PRISMA diagram (figure 1). Our electronic searches retrieved 966 records. Searching of other resources produced five additional references. After removing 66 duplicate references by endnote reference manager, we evaluated a total of 905 records, of which we excluded 893 based on the title and the abstract using Rayyan online site. The remaining 12 records were checked as full texts; three studies might be eligible as ongoing; further information is in the ongoing studies. We excluded three studies with reasons.

II. Risk of bias of included studies

Risk of bias within studies was assessed by Risk of bias tool for RCTs using *(Revman 5.4)* and by Newcastle Ottawa Scale (NOS) for non-randomized studies. According to our protocol, when a single domain was assessed at high or unclear risk, the trial was classified as being at high risk. As demonstrated in the risk of bias assessment (figures 2&3), we classified the four RCTs to be at overall high risk of bias. Authors' judgement with justification are shown in Supplementary File N.1. Two observational studies were assessed by NOS; one of them was of good quality *(Agrawal et al., 2014)* while the other study was of poor quality *(Taghaddosinejad et al., 2016)* (Table 4).

III. Effects of interventions

Primary outcomes

1. Mortality rate:

All the six included studies reported this outcome. Mortality was 41.37% (60/145 patients) in NAC treatment group and 60.28% (85/141 patients) in the control group. There was statistically significant difference between both groups favoring NAC group (OR = 0.38, 95% CI [0,23 to 0.66], P = 0.0005). Pooled studies were homogenous (Chi-square P = 0.52, I^2 = 0%).

By analyzing each subgroup according to NAC regimen separately, we found that both subgroups revealed a statistically significant difference favoring NAC group with (subtotal OR =0.46, 95% CI [0.24 to 0.86], P = 0.01) in the subgroup that received 21 h NAC regimen and (subtotal OR = 0.24, 95% CI [0,09 to 0.68], P = 0.007) in the 72 h NAC regimen subgroup. Pooled studies in each subgroup were homogenous. Intergroup difference was not significant (Chi-square P = 0.30, I^2 = 5.6%) (Figure 4).

We also did subgroup analysis according to type of studies (RCTs or OSs). The RCTs subgroup included four studies and revealed a statistically significant difference favoring NAC group (subtotal effect size 0.34, 95% CI [0.17 to 0.68], P = 0.002). In contrast, OSs subgroup included two studies and revealed a statistically non-significant difference between both groups (subtotal OR = 0.46, 95% CI [0.20 to 1.08], P = 0.08). Pooled studies in each subgroup were

homogenous. Intergroup difference was not significant (Chi-square P = 0.58, I^2 = 0%) (Figure 5).

2. Morbidity rate:

There were no clear data about morbidity in survivors in all studies.

Secondary outcomes

3. Duration of hospital stay in survivors:

Two RCTs reported this outcome. There was a statistically significant difference favoring NAC group (SMD = -1.73 days, 95% CI [-2.35, -01.11], P < 0.00001). Pooled studies were homogenous (Chi-square P = 0.23, I^2 = 31%).

We also did subgroup analysis according to NAC regimen. There was only one study in each subgroup, and both revealed statistically significant difference favoring NAC group with (SMD = -2.01 day, 95% CI [-2.78, -1.24], P <0.00001) in the subgroup that received the 21 h NAC regimen, and (SMD = -1.21 days, 95% CI [-2.26, -0.17], P = 0.02) in the subgroup that received the other regimen. Intergroup difference was not significant (Chi-square P = 0.23, I^2 = 30.8%) (Figure 6).

4. Duration of hospital stay in non survivors (survival time):

Three of the included studies reported this outcome. All studies used the same 21 hour NAC regimen. There was a statistically significant difference favoring NAC group (SMD = 0.87 day, 95% CI [0.37, 1.37], P = 0.0007). Pooled studies revealed significant heterogeneity (Chi-square P =< 0.00001, I^2 = 95%)

Subgroup analysis according to type of studies was done with two RCTs and one observational study. The RCTs subgroup revealed a statistically non-significant difference between both groups (SMD = 0.27day, 95% CI [-0.30, 0.84], P =0.35). Pooled studies revealed significant heterogeneity (Chi-square P=<0.00001, I^2=96%). While the OSs subgroup revealed a statistically significant difference favoring NAC group (SMD = 2.97 days, 95% CI [1.91, 4.04], P = 0.02). Intergroup difference was significant (Chi-square P <0.0001, I^2 = 94.8%) (Figure 7).

5. Need for mechanical ventilation:

Three RCTs of the included studies reported this outcome. About 35.82% (24/67 patients) were mechanically ventilated in the NAC treatment group versus 48.33% (29/60 patients) in the control group. There was a statistically non-significant difference between both groups (OR = 0.51, 95% CI [0,23 to 1.10], P=0.08). Pooled studies were with moderate heterogeneity (Chi-square P=0.18, I^2 =43)

By analyzing each subgroup according to NAC regimen separately, we found that the subgroup which received NAC with a dose of 300 mg/kg over 21 included one study and revealed a statistically non-significant difference between both groups (subtotal OR = 1.22, 95% CI [0.36 to 4.17], P=0.75). While the other subgroup revealed a statistically significant difference favoring NAC group (subtotal OR =0.26, 95% CI [0,09 to 0.76], P=0.01). Pooled studies in this subgroup were homogenous (Chi-square P= 0.77, I2=0%). Intergroup difference was significant (Chi-square P = 0.07, I^2 = 70.5 %) (Figure 8).

IV. Certainty of evidence

The certainty of evidence was mentioned in the methodology and summarized in the summary of findings for the four RCTs using GRADE criteria (GRADEpro GDT) online Guidelines Development Tool (Table 4).

Table (1): Baseline characteristics for populations of included studies

Study ID	Type	Group	Number	Male gender(%)	Mean age in years ± SD	Mean arrival time in hours	Manner of poisoning		Direct cases(%)	Hypotension N(%)	Mean PH ± SD	Altered sensorium
							Suicidal(%)	Accidental (%)				
Agrawal et al., 2014	OS	NAC	24	70.80%	27.74±8.86	.....	.....	.....	54.20%	20 (83.3%)	7.2±0.2	4.20%
		Control	22	72.70%		.....	.....	.....	68.20%	20 (90.9%)	7.2±0.2	9.10%
Bahalla et al., 2017	RCT	NAC	24	79%	64%<30 y	(54%) 3h	.....	.....	45.83%	24 (100%)	7.22±0.09	20.83%
		Placebo	26	61.50%			.....	.....	38.46%	26 (100%)	7.23±0.07	26.92%
El-ebiary and Abuelfad, 2017	RCT	NAC	15	46.70%	24.3±3.75	(1.5-6) h	86.70%	13.30%	.....	11 (73.3%)	7.4±0.1	.....
		Control	15	40.00%	26.3±7.36	(1-4.5) h	100%	0%	.....	14 (93.3%)	7.3±0.1	.....
Emam et al., 2020	RCT	NAC	30	36.7%	24.4±10.55	1.25±0.61	100%	0%	100%	27 (45%)	7.27±0.14	.....
		Control	30	36.7%	24.43±9.66	1.18±5.79	100%	0%	100%		7.28±0.13	.....
Taghaddosinejad et al., 2016	OS	NAC	30	30.40%	26.65±1.06	.....	.....	.....	.....	.....	.....	.....
		Control	33	56.50%	28.39±1.11	.....	.....	.....	.....	.....	.....	.....
Tehrani et al.,2013	RCT	NAC	22	50%	23.5±7.8	.....	100%	0%	100%	20 (90.9%)	7.36±0.13	27.30%
		Control	15	53.30%	24.7±6.4	.....	100%	0%	100%	15 (100%)	7.35±0.08	26.70%

Table (2): Summary of the included studies

Study ID	Design	Population	Sample	Dose of NAC	Results

			size	and Duration of treatment	
Agrawal et al., 2014	A cohort study.	Patients with acute AlP poisoning	46	300 mg/kg IV over 21 h	NAC along with supportive treatment might have improved survival in AlP poisoning.
Bahalla et al., 2017	Prospective intervention study (pilot study).(RCT)	Patients with severe AlP poisoning	50	300 mg/kg IV over 21 h	Antioxidant therapy in the form of NAC in sever AlP poisoning did not confer any survival benefit.
El-ebiary and Abuelfad, 2017	Randomized clinical trial.	Patients with acute AlP poisoning	30	1.33 g/kg IV over 72 h	NAC might be promising adjuvant therapy in treatment of acute AlP poisoning. Mortality rate and dopamine dose reduced in group received NAC.
Emam et al., 2020	Randomized clinical trial.	Patients with acute AlP poisoning	60	300 mg/kg IV over 21 h	Early administration of high doses of NAC along with adequate supportive treatment may have a survival benefit over supportive treatment alone.
Taghaddosinejad et al., 2016	Acase-control study	Patients with acute AlP poisoning	63	300 mg/kg IV over 20 h	The biochemical index of cardiotoxicity was found to elevate in both the case and control groups.
Tehrani et al.,2013	Randomized clinical trial.	Patients with acute AlP poisoning	37	1.33 g/kg IV over 72 h	NAC might have a therapeutic effect in acute AlP poisoning as it decreased mortality, mechanical ventilation, and duration of hospitalization in survivors

Table (3): Quality assessment of observational studies (Newcastle Ottawa Scale).

Study ID	Selection (max. 4)	Comparability (max. 2)	Exposure/outcome (max. 3)	Total (max.9)
Agrawal et al., 2014	4	2	3	9 Good quality
Taghaddosinejad et al., 2016	1	2	1	4 Poor quality

Table (4): Summary of findings and certainty of evidence for the four included randomized controlled trials

Outcomes	Anticipated absolute effects* (95% CI)		Relative effect (95% CI)	Number of participants (studies)	Certainty of the evidence (GRADE)
	Risk with [control]	Risk with [n-acetylcysteine]			
Mortality	66 per 100	**40 per 100** (25 to 57)	**OR 0.34** (0.17 to 0.68)	177 (4 RCTs)	Moderate
Comments: Our confidence in this result is moderate, downgraded one level for serious risk of bias (single RCT study at high risk of bias in allocation concealment and the three studies at high risk of bias in blindness of participants and personnel). N-acetylcysteine is likely to reduce mortality in population with acute aluminum phosphide poisoning.					
Morbidity	-- per --	**- per --** (-- to --)	not estimable	(0 studies)	-
Comments: This outcome was not clearly reported in the included studies					
Duration of hospital stay in survivors	-	SMD **1.73 SD fewer** (2.35 fewer to 1.11 fewer)	-	61 (2 RCT)	Moderate
Comments: Our confidence in this result is moderate, downgraded one level for serious risk of bias (the included study was with high risk of bias at both allocation concealment and blindness of participants and personnel). N-acetylcysteine is likely to reduce duration of hospital stay in acute aluminum phosphide survivors.					
Outcomes	**Anticipated absolute effects* (95% CI)**		**Relative effect (95% CI)**	**Number of participants (studies)**	**Certainty of the evidence (GRADE)**
	Risk with [control]	**Risk with [n-acetylcysteine]**			
Duration of hospital stay in non survivors	-	SMD **0.27 SD fewer** (0.3 fewer to 0.84 more)	-	63 (2 RCT)	Very Low[c]
Comments: Our confidence in this result is very low, downgraded one level for serious risk of bias (single study had unclear risk if bias at allocation concealment and selective reporting and high risk of bias at blindness of participants and personnel), downgraded one level for serious imprecision (wide confidence intervals crossing the line of no effect), and downgraded one level due to serious inconsistency (heterogeneity between studies p <0.00001). N-acetylcysteine may have no effect on survival time in patients expired after acute aluminium phosphide poisoning.					
Mechanical ventilation	48 per 100	**32 per 100** (18 to 51)	**OR 0.51** (0.23 to 1.10)	127 (3 RCTs)	Moderate[d]
Comments: Our confidence in this result is moderate, downgraded one level for serious risk of bias (one study had high risk of bias at allocation concealment and two studies at high risk of bias in blindness of participants and personnel). N-acetylcysteine may have no effect on need for mechanical ventilation in population with acute aluminum phosphide poisoning.					

The risk in the intervention group (and its 95% confidence interval) is based on the assumed risk in the comparison group and the relative effect of the intervention (and its 95% CI), CI: confidence interval; OR: odds ratio; SMD: standardized mean difference

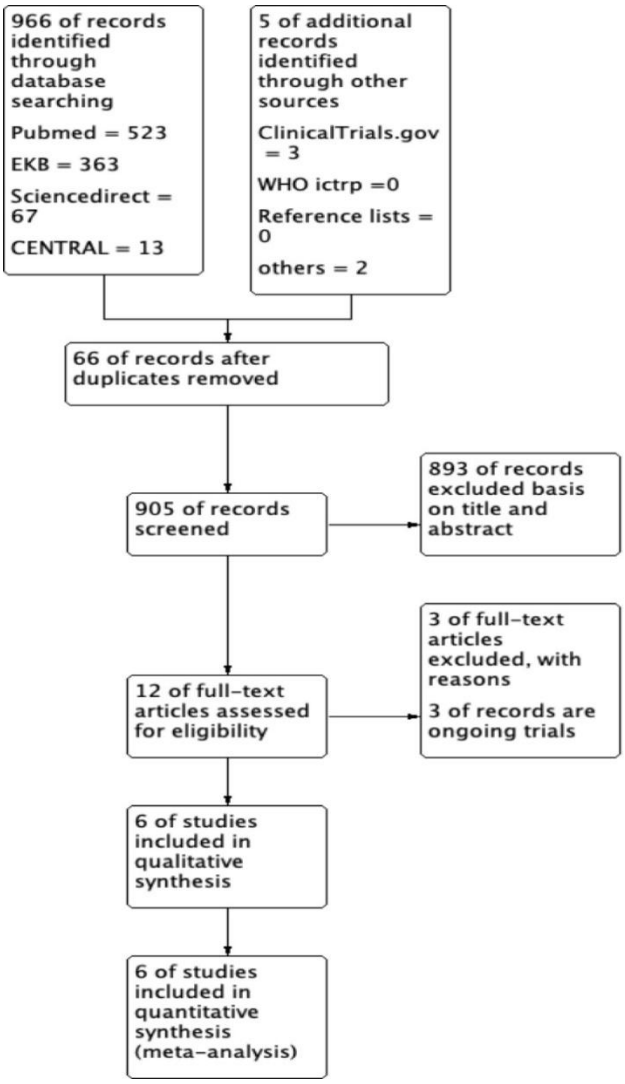


Figure 1: Study's PRISMA flow diagram. Date of search 19 June 2022.

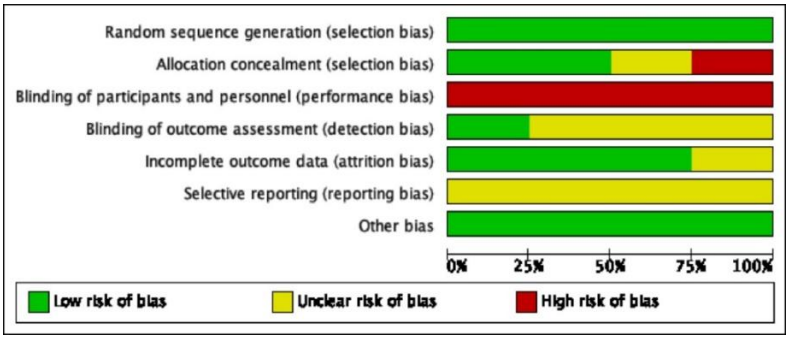


Figure 2: Risk of bias graph of included randomized controlled trials using the Cochrane risk of bias tool.

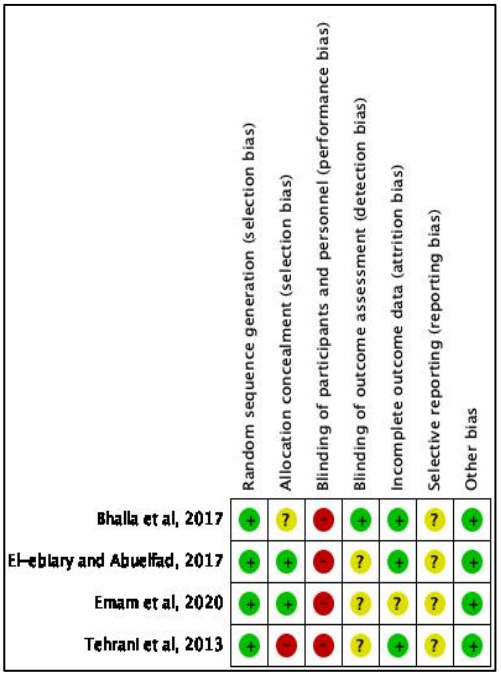
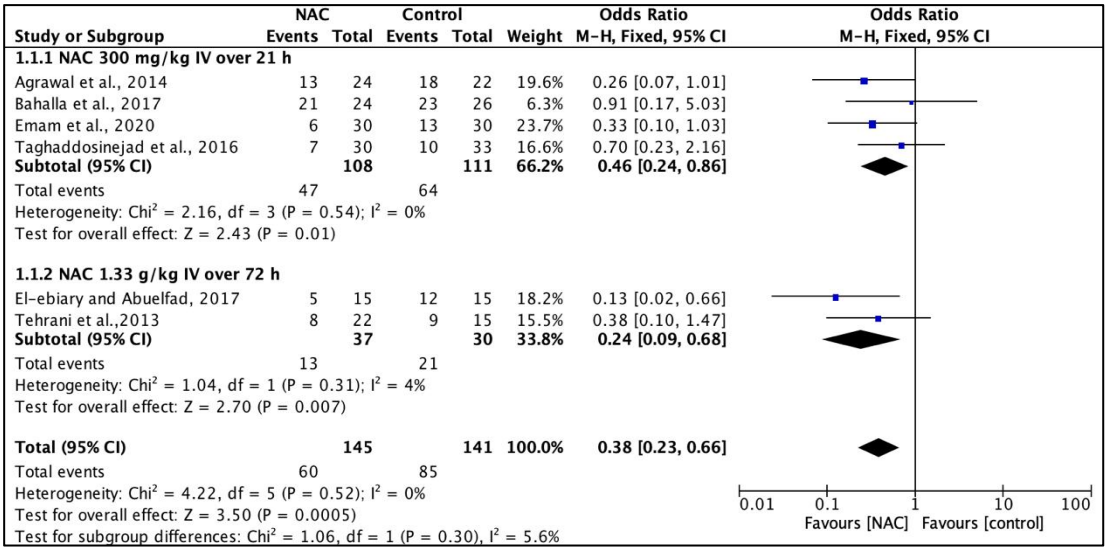
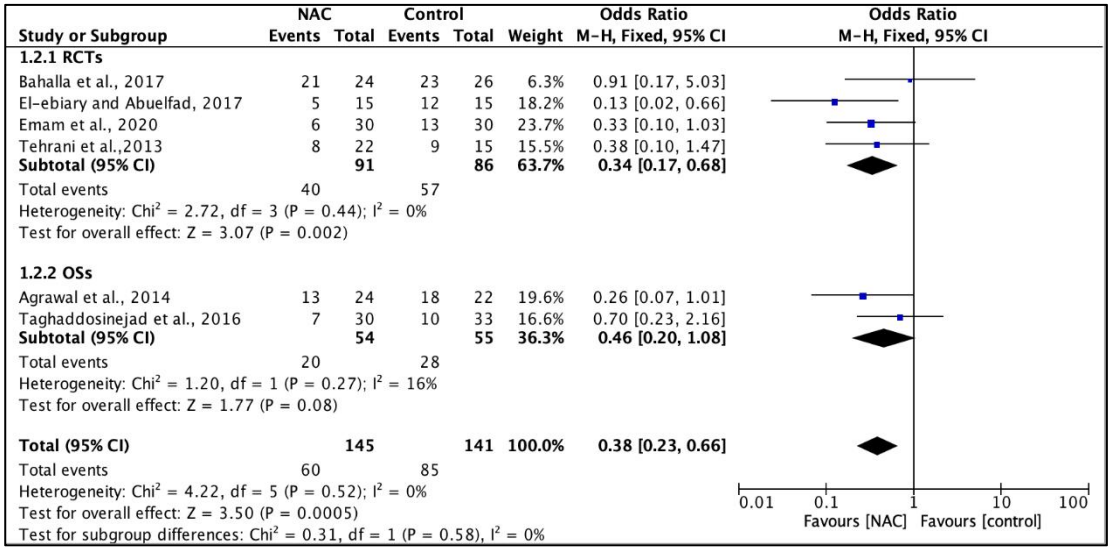


Figure 3: Risk of bias summary of included randomized controlled trials using the Cochrane risk of bias tool.



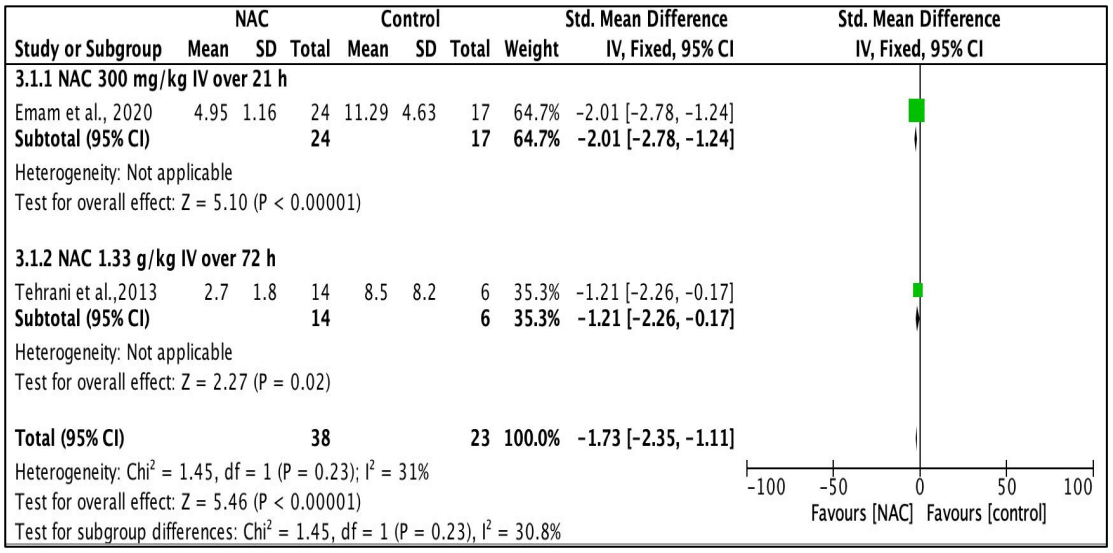
NAC = N-acetylcysteine, IV = intravenous, 95% CI = 95% confidence interval, M-H = Mantel–Haenszel method, Fixed = fixed effects model, Chi2 = Cochrane Q square test, df = degree of freedom, I2 = I squared test.

Figure 4: Forest plot showing the difference between NAC and control groups as regards mortality rate with subgroup analysis according to NAC regimens.



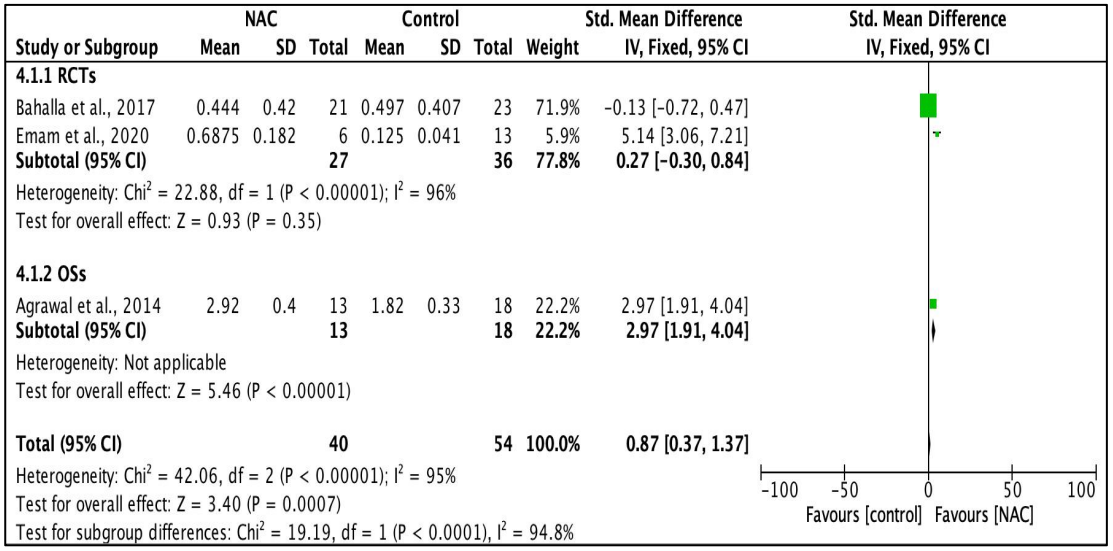
NAC = N-acetylcysteine, 95% CI = 95% confidence interval, M-H = Mantel–Haenszel method, Fixed = fixed effects model, Chi2 = Cochrane Q square test, df = degree of freedom, I2 = I squared test.

Figure 5: Forest plot showing the difference between NAC and control groups as regards mortality rate with subgroup analysis according to type of studies.



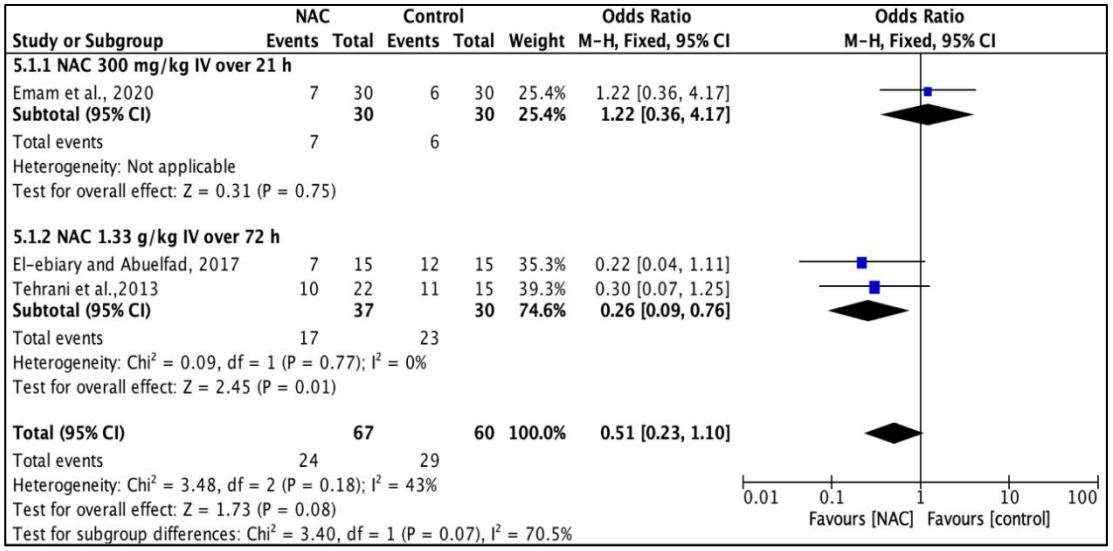
NAC = N-acetylcysteine, SD = standard deviation, 95% CI = 95% confidence interval, Std = standard, IV = inverse variance method, Fixed = fixed effects model, Chi2 = Cochrane Q square test, df = degree of freedom, I2 = I squared test.

Figure 6: Forest plot showing the difference between NAC and control groups as regards duration of hospital stay in survivors with subgroup analysis according to NAC regimens.



NAC = N-acetylcysteine, SD = standard deviation, 95% CI = 95% confidence interval, Std = standard, IV = inverse variance method, Fixed = fixed effects model, Chi2 = Cochrane Q square test, df = degree of freedom, I2 = I squared test.

Figure 7: Forest plot showing the difference between NAC and control groups as regards duration of hospital stay in non survivors (survival time).



NAC = N-acetylcysteine, 95% CI = 95% confidence interval, M-H = Mantel–Haenszel method, Fixed = fixed effects model, Chi2 = cochrane Q square test, df = degree of freedom, I2 = I squared test.

Figure 8: Forest plot showing the difference between NAC and control groups as regards the need for mechanical ventilation with subgroup analysis according to NAC regimen.

Discussion

A. Summary of main results

This systematic review of N-acetylcysteine usage in acute aluminium phosphide poisoning included six studies: four RCTs and two OSs with total 286 participants.

The overall meta-analysis found that N-acetylcysteine could reduce the mortality rate which was reported in the six included studies and hospital stay duration in survivors which was reported in two of the included studies with a statistically significant difference.

Although quantitative analysis of duration of hospital stay in non survivors (survival time) which was reported in three studies showed significant prolongation, we could not consider meta-analysis because of significant unresolved heterogeneity between studies (Chi-square P =< 0.00001, I^2 = 95%). The cause of heterogeneity may be due to the difference between the included participants in each study as Bhalla *et al. (2017)* included only patients with severe toxicity manifested by hypotension and shock which would affect the survival time. Another

reason may be due to difference in type of studies, two were RCTs and one was a cohort observational study.

Meta-analysis for the difference between NAC and control groups as regards need for mechanical ventilation revealed that NAC did not affect this outcome. Pooled studies were with moderate heterogeneity (Chi-square P = 0.18, I^2 = 43%). This heterogeneity was resolved by subgroup analysis according to NAC regimen.

Our subgroup analyses according to NAC regimens showed that 300 mg/kg IV NAC over 21 h (300 mg/kg over 20 h or 150 mg/kg over one hour then 50 mg/kg over four hours then 100 mg/kg over 16 hours) significantly decreased both mortality rates and duration of hospital stay in survivors. On the other hand, it significantly increased the survival time in non survivors, but there were significant heterogeneity (Chi-square P =< 0.00001, I^2 = 95%).

While the other regimen of 1.33 g/kg IV NAC over 72 h regimen (140 mg/kg as a loading dose then 70 mg/kg every 4 h up to 17 doses) significantly reduced each of the following: the mortality rate, duration of hospital stay in survivors, and the need for mechanical ventilation

There was no difference between the two NAC regimens as regards reduction of both the mortality rate and duration of hospital stay in survivors.

Our subgroup analysis according to type of studies revealed that RCTs subgroup showed that NAC usage resulted in a significant reduction in mortality rate and hospital stay duration in survivors but did not affect both survival time and the need for mechanical ventilation.

While OSs subgroup showed that NAC usage resulted in a significant increase in survival time in non survivors, it did not affect the mortality rate.

B. Quality of the evidence

The certainty of evidence (quality of evidence) of the four included RCTs was summarized in the summary of evidence and as follows:

Regarding primary outcomes, the certainty of evidence for the mortality rate was moderate, it was downgraded one level due to serious risk of bias of the included studies. While certainty of evidence for morbidity rate was not reported.

Regarding secondary outcomes, the certainty of evidence for duration of hospital stay in survivors was moderate, it was downgraded one level due to serious risk of bias of the included study. The certainty of evidence for duration of hospital stay in non survivors was very low, it was downgraded one level due to serious imprecision and one level for serious inconsistency. Finally, the certainty of evidence for mechanical ventilation was moderate, it was downgraded one level due to serious risk of bias.

C. Potential biases in the review process

We performed this review according to a predefined protocol, following guidance from the *Cochrane Handbook for Systematic Reviews of* Interventions, which we completed and published prior to beginning of the review process. We used a comprehensive search strategy to minimize possible publication bias. It is unlikely that this strategy missed any published studies or large unpublished studies. We could not formally evaluate publication bias due to the small number of trials identified.

We included both randomized clinical trials and observational studies to identify as large as possible data published on our topic, and this is one of the limitations in our study. Two studies (40%) are observational studies, one of them of poor quality and observation time was 24 hours from admission *(Taghaddosonijad et al., 2016).*

To overcome this limitation, subgroup analyses of RCTs alone were done and we created summary of findings (certainty of evidence) for only the four included RCTs.

Another limitation was different NAC regimens and doses. To overcome this issue, subgroup analyses according to NAC regimen were done.

The last detected limitation was significant, unresolved heterogeneity between studies reported duration of hospital stay in non survivors (survival time).

D. Agreement and disagreement with other studies or reviews

This study is the first systematic review and meta-analysis done in this topic.

A randomised controlled trial published after June 2022 (our search limit) was done by *Ashraf and his colleagues* to determine the effect of NAC on mortality rate in AlP acutely intoxicated patients. It was conducted in Lahore, Pakistan with 96 participants; 48 in each group (NAC and control). The study revealed a significant reduction in mortality rate favoring NAC group (p =0.024) which agreed with our study (P =0.0005) *(Ashraf et al., 2022).*

Recent systematic review and meta-analysis was done by Rashid *and his colleagues* about NAC use in rodenticide poisoning including yellow phosphorous, zinc phoshsphide, aluminium phosphide and others *(Rashid et al., 2022).*

Mortality in *Rashid et al., 2022* study showed that meta-analysis of RCTs (OR: 0.25; 95% CI: 0.11-0.59; n = 2) and retrospective studies (OR: 0.34; 95% CI: 0.15-0.78; n = 3) showed a significant reduction in mortality, whereas pooled analysis of prospective studies recorded a non-significant effect. And thus, agreed with our study which showed significant reduction in that outcome in meta-analysis of RCTs but non-significant reduction in meta-analysis of OSs.

Unlike our study *Rashid et al., 2022* showed a significant reduction of intubation or ventilation (OR: 0.25; 95% CI: 0.11-0.60; 2 RCTs) and a non-significant reduction in duration of hospital stay (P = 0.41) between patients who received NAC and who were not treated with NAC. This study is not similar to ours, as rodenticides are many types of different mechanisms of toxicity, but we discussed it because aluminium phosphide is one of the rodenticides.

Conclusion and Recommendations

N-acetylcysteine in treatment of acute aluminium phosphide poisoning can reduce the mortality rate and duration of hospital stay in survivors, in addition to increase survival time in non-survivors. We recommend the use of NAC in acute aluminium phosphide poisoning at two different regimens that were mentioned in our study. Further high quality randomized controlled trials targeting a broader population are recommended. We also recommend not to use the Egyptian Knowledge Bank (EKB) in the advanced search, as its results are not reproducible.

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استخدام ان-اسيتيل سيستايين في علاج التسمم الحاد بفوسفيد الألومنيوم: مراجعة منهجية وتحليل تلوي

سلوى عبد المنعم إبراهيم حسين[1] و ايناس ابوالوفا التفتازاني[3] و عبير محمد إسماعيل هجرس[2] و هند الهلالي[3] و ولاء جمعة عبد الحميد[3]

الملخص العربي

الخلفية: فوسفيد الألومنيوم (AlP) هو مبيد قوارض شائع الاستخدام يمنع الفسفرة المؤكسدة، كما يتسبب في نضوب الجلوتاثيون مما يؤدي إلى خلل في جدار الخلية. يعتبر ان–اسيتيل سيستايين من سلائف الجلوتاثيون ويمكن أن يتفاعل مع أنواع الاكسجين التفاعلية (ROS) ولذلك سيكون فعالاً في علاج التسمم الناتج عن فوسفيد الألومنيوم. **الهدف من العمل:** تقديم مراجعة منهجية قائمة على الأدلة حول دور ان–اسيتيل سيستايين في علاج التسمم الحاد بفوسفيد الالومنيوم والذي قد يساعد في وضع مبادئ توجيهية واضحة لعلاج التسمم بمثل هذا السم القاتل. **المنهجية:** اتبعنا إرشادات PRISMA أثناء إعداد هذه الدراسة. تم البحث في PubMed وEKB وScienceDirect و Cochrane Centralلتحديد المؤلفات المنشورة من البداية حتى يونيو 2022. بالإضافة إلى ذلك، بحثنا عن الدراسات الجارية وقوائم المراجع لدراسات مشابهة. وقد قمنا بتضمين التجارب العشوائية المنتظمة والدراسات القائمة على الملاحظة المنشورة باللغة الإنجليزية تلك التي تنطبق عليها معايير الاشتمال. **النتائج:** تضمنت هذه المراجعة المنهجية أربعة تجارب عشوائية منتظمة ودراستين قائمتين على الملاحظة بإجمالي 286 مشاركًا. وقد كشفت أن هناك انخفاضًا ملحوظاً في معدل الوفيات ومدة الإقامة في المستشفى للناجين. بالإضافة الى زيادة ملحوظة في وقت البقاء على قيد الحياة لدى غير الناجين في المرضى الذين تلقوا العقار مقارنة مع أولئك الذين لم يتلقوه. كما كشفت عن عدم وجود فرق ملحوظ بين المجموعتين فيما يتعلق بالحاجة إلى التهوية الميكانيكية. **الخلاصة:** استخدام ان–اسيتيل سيستايين في علاج التسمم الحاد بفوسفيد الالومنيوم يمكن أن يقلل من معدل الوفيات ومدة الإقامة في المستشفى لدى الناجين ويزيد من وقت البقاء على قيد الحياة في الذين لم ينجوا.

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