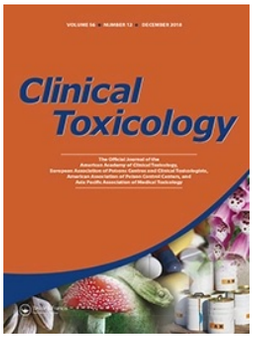

Clinical Toxicology

ISSN: 1556-3650 (Print) 1556-9519 (Online) Journal homepage: https://www.tandfonline.com/loi/ictx20

N-acetylcysteine as a treatment for amatoxin poisoning: a systematic review

Jiaming Liu, Yang Chen, Yanxia Gao, Joseph Harold Walline, Xin Lu, Shiyuan Yu, Lina Zhao, Zengzheng Ge & Yi Li

To cite this article: Jiaming Liu, Yang Chen, Yanxia Gao, Joseph Harold Walline, Xin Lu, Shiyuan Yu, Lina Zhao, Zengzheng Ge & Yi Li (2020): *N*-acetylcysteine as a treatment for amatoxin poisoning: a systematic review, Clinical Toxicology, DOI: 10.1080/15563650.2020.1784428

To link to this article: https://doi.org/10.1080/15563650.2020.1784428

Published online: 01 Jul 2020.

CLINICAL TOXICOLOGY
https://doi.org/10.1080/15563650.2020.1784428

Taylor & Francis
Taylor & Francis Group

REVIEW

N-acetylcysteine as a treatment for amatoxin poisoning: a systematic review

Jiaming Liu[a], Yang Chen[b], Yanxia Gao[c], Joseph Harold Walline[d], Xin Lu[a], Shiyuan Yu[a], Lina Zhao[a], Zengzheng Ge[a] and Yi Li[a]

[a]Department of Emergency Medicine, Peking Union Medical College Hospital, Chinese Academy of Medical Sciences & Peking Union Medical College, Beijing, People's Republic of China; [b]Department of Rheumatology and Clinical Immunology, Peking Union Medical College Hospital, Chinese Academy of Medical Sciences & Peking Union Medical College, Beijing, People's Republic of China; [c]Department of Emergency Medicine, The First Affiliated Hospital of Zhengzhou University, Zhengzhou, People's Republic of China; [d]Accident and Emergency Medicine Academic Unit, Prince of Wales Hospital, The Chinese University of Hong Kong, Hong Kong, People's Republic of China

ABSTRACT

Introduction: Amatoxin leads to the majority of deaths by mushroom poisoning around the world. Amatoxin causes gastrointestinal disturbances and multiple organ dysfunction, including liver and renal failure. As a potential treatment for amatoxin poisoning, *N*-acetylcysteine (NAC) has been used for decades but its benefit is still unproven.

Objectives: We undertook a systematic review to evaluate the performance and safety of *N*-acetylcysteine on patients suffering amatoxin intoxication.

Methods: We searched Pubmed, EMBASE, CENTRAL and SinoMed databases, from inception to August 31, 2019. Articles were eligible if there were five or more patients with amatoxin poisoning and *N*-acetylcysteine was included in the therapeutic regimen. Mortality rate including liver transplant cases (MRLTi) was the primary outcome. Mortality rate not including liver transplant cases, liver and renal function, clinical complications, as well as any adverse reactions to intravenous NAC were secondary outcomes.

Results: Thirteen studies with a total of 506 patients were included. The MRLTi of amatoxin-poisoning patients with NAC treatment was 11% (57/506), and a MRLTe of 7.9% (40/506) and a liver transplantation rate of 4.3% (22/506). Transaminase concentrations generally peaked around 3 days after ingestion, prothrombin time/International Normalized Ratio (PT/INR) generally worsened during the first 3–4 days after ingestion before returning to normal four to 7 days after ingestion, and Factor V levels normalized in about 4–5 days after ingestion in patients treated with NAC. Renal failure was reported in 3% (3/101) and acute kidney injury was reported in 19% (5/27). Gastrointestinal bleeding occurred in 21% (15/71). Anaphylactoid reactions were the principle adverse reaction to NAC treatment in amatoxin-poisoning patients with an incidence of 5% (4/73).

Conclusions: NAC treatment combined with other therapies appears to be beneficial and safe in patients with amatoxin poisoning. Until further data emerge, it is reasonable to use NAC in addition to other treatments for amatoxin poisoning.

ARTICLE HISTORY
Received 18 December 2019
Revised 8 June 2020
Accepted 11 June 2020

KEYWORDS
Amatoxin; mushroom poisoning; acetylcysteine; liver failure; liver transplantation; renal insufficiency

Introduction

Among mushroom-derived toxins, amatoxin accounts for more than 95% of deaths related to mushroom poisoning [1]. Amatoxin is a potent poison that can non-covalently bind and inhibit RNA polymerase II activity, causing a reduction in cellular mRNA and protein synthesis (especially in liver cells [2]), finally leading to cell death [3]. Amatoxin also induces the formation of reactive oxygen species which can cause oxidative stress-related cellular damage, leading to apoptosis *via* an interaction with TNF-α or p53 [4].

Despite a high mortality rate of between 5% and 35% in amatoxin-poisoning patients [5], there is no confirmed antidote to reverse the toxicity of amatoxin [6]. Multiple-Dose Activated Charcoal, intravenous penicillin, intravenous or oral silymarin/silibinin, extracorporeal elimination methods, and liver transplantation constitute the current bundle of potential treatments [7].

N-acetylcysteine (NAC), which is hepatoprotective in paracetamol poisoning [8], is also a potential treatment of amatoxin intoxication that has been used to treat mushroom poisoning since the early 1990s [7,9]. The first case series of intravenous NAC treatment in amatoxin-poisoning patients showed a lower mortality rate (8.22%) than previous studies and therefore suggested a potential role for NAC in treating amatoxin intoxication [10]. However, controversial results with NAC were seen in both animal experiments and clinical cases in later years [11,12]. Whether NAC should be routinely used in patients with amatoxin intoxication remains an unsettled question.

CONTACT Yi Li billliyi@126.com Department of Emergency Medicine, Peking Union Medical College Hospital, Chinese Academy of Medical Sciences & Peking Union Medical College, Beijing, People's Republic of China

In this article, we performed a systematic review to assess the possible therapeutic effects of NAC on amatoxin poisoning patients' mortality and organ functions.

Methods

Search strategy

We conducted electronic searches of Pubmed, EMBASE, Cochrane Central Register of Controlled Trials (CENTRAL), and the Chinese Biomedical Literature Database (SinoMed, in Chinese) from their inception to August 31, 2019. Search terms included "acetylcysteine," "N-acetylcysteine," "N-acetyl-L-cysteine," "NAC," "amatoxins," "amanitins," "mycotoxins," "alpha-amanitin," "amanita," "basidiomycota," "agaricales," "mushroom poisoning," "mycotoxicosis," "death cap," "destroying angel," and "hepatotoxic mushrooms." Additionally, we also searched the reference lists in any included studies.

Eligibility criteria

We included articles in this systematic review that were case series, cohort studies or clinical trials with five or more patients. Amatoxin poisoning was diagnosed by one of the following criteria: (1) confirmed detectable amanitin in patients' blood or urine; (2) a remnant of food consumed by patients was confirmed to be amatoxin-containing mushrooms; (3) a history of mushroom consumption and typical clinical manifestations of amatoxin poisoning (e.g., vomiting, diarrhea, and transaminase elevation). The intervention described in the included articles had to include NAC, regardless of dose, dosage form, duration or formula. We excluded non-human studies and limited our search to articles in either English or Chinese.

Outcome measures

The primary outcome was mortality rate including liver transplant cases (MRLTi) because both death and liver transplant are considered poor outcomes in cases of amatoxin poisoning.

MRLTi was calculated as follows:

$$\text{MRLTi} = \frac{\text{number of deaths} + \text{number of LTs} - \text{number of patients who died after LT}}{\text{number of treated patients}}$$

MRLTi: mortality rate including liver transplant cases; LT: liver transplant.

Secondary outcomes included the mortality rate excluding liver transplant cases (MRLTe). MRLTe was calculated as the number of deaths divided by the number of treated patients. Additional secondary outcomes included liver function [including transaminase, bilirubin, prothrombin time/International Normalized Ratio (PT/INR), and Factor V levels, as well as any occurrence of hepatic encephalopathy], renal function (including elevated creatinine levels and any occurrence of renal insufficiency or renal failure), and any occurrence of complications in the clinical course or adverse reactions of intravenous NAC.

Data extraction

We used a standardized data extraction form to collect the following data from each study: (1) General information: study ID, title, first author, and year of publication; (2) Study characteristics: study type; (3) Participants: average age, diagnostic criteria, number of patients, and interval between ingestion and admission to hospital; (4) Interventions: interventions used in treatment; dose, dosage form, duration, and formulation of NAC treatment; (5) Outcomes: MRLTi, indicators of liver and renal function, and any complications in the clinical course or adverse reactions from intravenous NAC (Table 1).

Results

The literature search from the electronic databases identified 213 records (211 records in English and 2 records in Chinese). Thirteen studies met our inclusion criteria (eight case reports with $N > 5$ [10,13–19] and five retrospective cohort studies [12,20–23]). A total of 506 patients from these 13 studies were included in our final analysis (Figure 1). Among the included articles, sample sizes ranged from 5 cases [17] to 157 cases [15].

One study reported pediatric patients with mean age of 6 years old [17]. One study distinguished pediatric patients from adult patients, with mean ages of 15 and 34 years, respectively [13]. The other studies did not separate pediatric from adult patients, and reported mean ages from 32 [23] to 52 years [15]. In five studies, the diagnosis of amatoxin poisoning included laboratory confirmation of amatoxin in patients' blood, urine, or food remnants [14,15,17,20,22]. In another seven studies, the diagnosis rested upon a history of mushroom consumption with clinical findings typical of amatoxin poisoning [12,13,16,18,19,21,23]. The last study did not specify the criteria used in arriving at the diagnosis of amatoxin poisoning [10].

Only some of the patients in five studies received NAC treatment [12,13,17,19,21]. MRLTi and MRLTe were calculated only from patients treated with NAC in two of these studies [12,17] and from all patients in the other three studies [13,19,21]. Indicators of liver and renal function, complications or adverse reactions to intravenous NAC, and the interval between ingestion and admission to hospital were collected from only NAC treated patients in one of the

Table 1. Reports of amatoxin poisoning treated with *N*-acetylcysteine.

Authors, Publication Year [Ref.]	Country	Study Type	Number of Cases/ Average Age	Diagnostic Criteria/ Inclusion Criteria	Formula of NAC Treatment	Adverse reaction/ Complication (Number of cases)	Other Treatment (Number of patients received)	Time from ingestion to admission to hospital	Average Length of Hospital Stay	MRLTi	MRLTe
Locatelli et al., 1992 [10]	Italy	Case series	73 cases 2–84 years old	–	Intravenous 150 mg/kg bolus followed by 50 mg/kg every four hours for prolonged time (between 3 and 18 days)	Anaphylactoid reactions (4)	Forced diuresis (73)	–	–	8% (6/73)	8% (6/73)
Yamada et al., 1998 [13]	USA	Case series	10 cases (NAC: 8 cases) 23 (12–68) years old	Gastrointestinal symptoms beginning ≥6 hours after consuming wild mushrooms	–	–	H2-blockers (10); Activated charcoal (9); Penicillin (6); Fresh frozen plasma (3); Vitamin K (6);	28h in average (16.5h–48h) (to start of medical care)	4.4 days (2–8 days)	20% (2/10)	20% (2/10)
Montanini et al., 1999 [14]	Italy	Case series	11 cases 38.5 (5–72) years old	Gastrointestinal symptoms + Food remnant identification/ HPLC detection of α-amanitin	Intravenous 200 mg/kg bolus followed by 10 mg/kg/h until the screened values returned to normal. 2.5 mg/kg/h during the entire ICU staying.	–	Venovenous haemodiafiltration (11); Laxatives (hydrated magnesium sulphate) (11); Activated charcoal (11); Penicillin (11);	6h–13h	9.6 days (8 – 16 days) (ICU stay)	9% (1/11)	0% (0/11)
Escudie et al., 2007 [19]	France	Case series	27 cases (NAC: 22 cases) 48 ± 17 years old	Mushroom ingestion history + gastrointestinal symptoms + transaminase elevation + identification of mushroom by experts	Intravenous 100 mg/kg every 16h	–	Silibinin (3) Penicillin (15) Activated charcoal (15) Hemodialysis or continuous hemodiafiltration (6)	32h in average	–	30% (8/27)	22 % (6/27)
Locatelli et al., 2010 [15]	Italy	Case series	157 cases 51.9 ± 18.5 years old	Mushroom ingestion history+ α-amanitin toxic levels (>10 ng/ml RIA, >1.5 ng/ml EMIT)	Intravenous 150mg/kg bolus followed by 300mg/kg/day until 48h after mushroom ingestion in patients without hepatitis and as long as ALT < 200 UI/L in patients with hepatic damage (performed for 2–21 days)	No adverse reactions	Forced diuresis; Activated charcoal gastrointestinal dialysis; (not known)	12h–168h (to start of NAC treatment)	–	2.5% (4/157)	1.9% (3/157)
Bergis et al., 2012 [20]	Germany	Retrospective cohort study	20 cases 48.2 years old	Urinary amanitin Toxin > 1.5ng/ml	Intravenous 150mg/kg bolus followed by 50mg/kg within 4 hours followed by 100mg/kg within 16 hours	Renal failure (1)	Activated charcoal (20); Silibinin (20); FPSA (9);	37.5h in average	9.6 days	5% (1/20)	5% (1/20)
Ahishali et al., 2012 [16]	Turkey	Case series	77 cases 41.94 ± 15.40 years old	Gastrointestinal symptoms beginning ≤24 hours after consuming wild mushrooms	Intravenous 210 mg/kg/day for first 3–5 days	Renal failure (1) Acute liver failure (7) No adverse reactions	Gastric lavage (unknown); Activated charcoal (77); Penicillin (77); Silymarin (75); Hemofiltration (77);	12.9h in average	4.4 days	3% (2/77)	3% (2/77)
Grabhorn et al., 2013 [17]	Germany	Case series	5 cases (NAC: 4 cases) 6 (1–10 years old)	α-amanitin detection in food remnant or urine/ Fulminant hepatic failure + Mushroom ingestion history	Intravenous 100 mg/kg/day	Renal failure (1)	Silibinin (4); Activated charcoal (4); Vitamin K (4); Laxatives (lactulose) (4); Penicillin (1); Plasmapheresis (1);	11.3h in average (10–13h)	–	25% (1/4)	0% (0/4)

(*continued*)

Table 1. Continued.

Authors, Publication Year [Ref.]	Country	Study Type	Number of Cases/ Average Age	Diagnostic Criteria/ Inclusion Criteria	Formula of NAC Treatment	Adverse reaction/ Complication (Number of cases)	Other Treatment (Number of patients received)	Time from ingestion to admission to hospital	Average Length of Hospital Stay	MRLTi	MRLTe
Akın et al., 2013 [23]	Turkey	Retrospective cohort study	NAC group: 24 cases, 32.0 ± 15.4 years old; Control group: 16 cases, 34.0 ± 16.5 years old.	Clinical presentation and hepatotoxicity consistent with A. phalloides intoxication	Intravenous 12 g/day in four divided doses	–	Gastric enemas (40); Hemoperfusion (40); Penicillin (40); Activated charcoal (40); Laxatives (lactulose) (40);	NAC group: 8.3h in average; Control group: 10.0h in average	–	4% (1/24)	4% (1/24)
Karvellas et al., 2016 [21]	USA	Retrospective cohort study	18 cases (NAC: 16 cases) 49 years old	ALF or ALI caused by A. phalloides	Intravenous 140 mg/kg bolus followed by 70 mg/kg for 17 h then continued at the same concentration at the discretion of the clinician. (performed for 4 days in average)	Arrhythmia (4); GI bleeding (3); Tracheal infection (2)	Silibinin (7); Activated charcoal (6); Penicillin (6); Nasobiliary drainage (1); Haemoperfusion or MARS (7)		–	44% (8/18)	11% (2/18)
Bonacini et al., 2017 [18]	USA	Case series	27 cases 47 (15–82) years old	Mushroom ingestion history + Transaminase levels 10x over the upper limit of normal (>400 IU/L).	Intravenous 140 mg/kg bolus followed by 70 mg/kg every 4 hours	Acute renal impairment (5)	Activated charcoal (27); Penicillin (19); Silymarin (14); Continuous venovenous hemofiltration (4);	<24h for all	–	15% (4/27)	11% (3/27)
Trakulsrichai et al., 2017 [12]	Thailand	Retrospective cohort study	55 cases (NAC: 35 cases) 43.85 ± 21.65 years old	Gastrointestinal symptoms beginning >5 hours after consuming wild mushrooms or mushroom ingestion history + Transaminase elevation	Intravenous	AKI (25/54); GI bleeding (12/53)[a]	Activated charcoal (27); Penicillin (18); Silymarin (8);	Not known in patients with NAC treatment	4 days	34% (12/35)	34% (12/35)
Kieslichova et al., 2018 [22]	Czech Republic	Retrospective cohort study	23 cases 49.22 ± 17.52 years old	Mushroom ingestion history + Gastrointestinal symptoms + Food remnant or feces identification	Intravenous		Activated charcoal (23); Silibinin (23); Hemoperfusion (1); Plasmapheresis (9); CRRT (6); FPSA (5);	26.3h in average	23.7 days (4–104 days)	30% (7/23)	9% (2/23)

NAC: *N*-acetylcysteine; MRLTi: Mortality rate including liver transplant cases; MRLTe: Mortality rate excluding liver transplant cases; FPSA: Fractionated plasma separation and absorption; MARS: Molecular adsorbent recirculating system; CRRT: Continuous renal replacement therapy.

[a]Means (number of cases/total number of cases recorded).

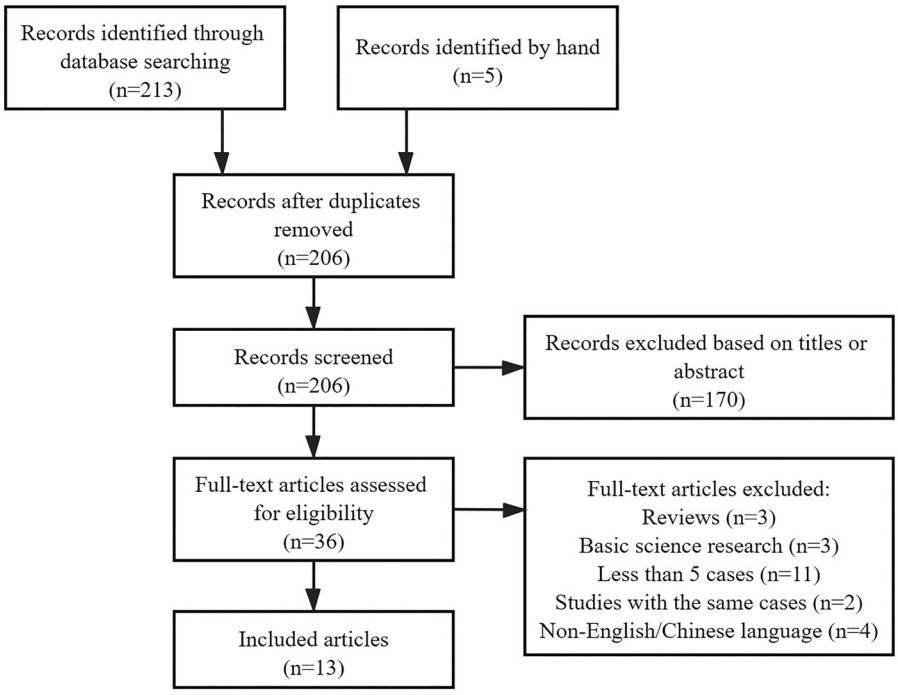

Figure 1. Flowchart of the literature search and study selection.

studies [17] and from all patients in the other four studies [12,13,19,21] as we could not distinguish this element from NAC treatment.

As for the intervention, ten studies had detailed descriptions of the dose, dosage form, duration and formula of NAC treatment [10,14–21,23], while complete information was not provided in three studies [12,13,22]. The most commonly used treatments besides NAC and general supportive measures included activated charcoal, penicillin, and silymarin/silibinin, which were reported to be used in twelve [12–23], nine [12–14,16–19,21,23], and eight [12,16–22] studies, respectively. Extracorporeal elimination methods were also used in nine studies, primarily in those patients with a short time interval between ingesting a mushroom and starting treatment [14,16–23]. The detailed intervention methods for each study are listed in Table 1.

Primary outcome

Mortality rate including liver transplant cases

The overall MRLTi was 11% (57 out of 506 cases) [10,12–23], although this ranged between 2.5% [15] and 44% [21]. As for pediatric patients younger than 18 years old, the MRLTi was 14% (1 out of 7 cases) in two studies [13,17]. Some children received NAC in two other studies that did not give detailed outcomes by age, so their MRLTi rate is unknown [10,14]. The interval between ingestion and admission to hospital ranged between 8.3 h [23] and 37.5 h [20]. The MRLTi in studies with a mean interval <24 h (early presenters) was 6.3% (9 out of 143 cases) [14,16–18,23], while the MRLTi in studies with mean interval >24 h (late presenters) was 23% (18 out of 80 cases) [13,19,20,22]. Three studies did not provide interval data [10,12,21], and they were not included in the calculation of early or late presenters.

Considering the effects of other treatments on amatoxin poisoning, we calculated the MRLTi of patients receiving combined treatments of NAC with silibinin/silymarin, penicillin G, and extracorporeal elimination methods. Among the studies included, silibinin/silymarin, penicillin G, and extracorporeal elimination methods were used in 154, 193, and 169 patients, respectively. The MRLTi of patients with both NAC and silibinin/silymarin treatments was 10.5% (15 out of 143 cases) [12,16–18,20,22], the MRLTi of patients with both NAC and penicillin treatments was 7.0% (9 out of 129 cases) [12,14,16,17,23], and the MRLTi of patients with both NAC and extracorporeal elimination treatments was 7.4% (10 out of 136 cases) [14,16,17,20,22,23].

Secondary outcomes

Mortality rate excluding liver transplant cases and the rate of liver transplantation

The overall MRLTe was 7.9% (40 deaths out of 506 cases), ranging from 0% [14,17] to 34% [12]. The overall rate of liver transplantation despite NAC treatment was 4.3% (22 out of 506 cases), ranging from 0% [10,13,20,23] to 33% [21].

Liver function

Transaminase levels, which may reflect the extent of damage to liver cells, was recorded on hospital admission in 213 patients [12,14,16,19,20,22]. The mean AST result on admission ranged from 117 IU/L [16] to 2,130 IU/L [22] (136 IU/L [14,16] in early presenters and 1822 IU/L [20,22] in late presenters), while the median AST ranged from 159.5 IU/L [12]

to 1,747 IU/L [22]. The mean ALT result on admission ranged from 111.7 IU/L [16] to 2,295 IU/L [22] (240 IU/L [14,16] in early presenters and 2160 IU/L [20,22] in late presenters), while the median ALT ranged from 122 IU/L [12] to 2,111 IU/L [22]. The peak transaminase value was attained at around 3 days after ingestion [13,14,16,18,20]. Among 124 patients with detailed documentation [12,13,17–19], the mean peak value of AST ranged from 2,923 IU/L [13] to 5,237 IU/L [17], while the median peak value of AST ranged from 871 IU/L [12] to 5,480 IU/L [18]. The mean peak value of ALT ranged from 4,138 IU/L [13] to 5,324 IU/L [19], and the median peak value of ALT ranged from 1,426 IU/L [12] to 6,282 IU/L [18]. Except for those studies that specifically included acute liver failure patients, the incidence of acute liver failure in amatoxin poisoning patients being treated with NAC was 9% (7 out of 77 cases) in one study [16]. Acute liver failure was also reported in two other studies, but its exact incidence was not provided [12,20].

Impaired liver function may lead to hyperbilirubinemia and hepatic encephalopathy due to a diminished detoxification capacity. In studies with detailed information, the mean bilirubin on admission ranged from 0.69 mg/dL to 2.81 mg/dL [16,19,20,22] (0.69 mg/dL [16] in early presenters and 1.84 mg/dL [19,20,22] in late presenters), while the median peak value of bilirubin ranged from 1.4 mg/dL to 5.6 mg/dL [12,17,18]. Seven studies recorded some degree of hepatic encephalopathy (55 cases). Cases of reported hepatic encephalopathy included grades I–II (8 patients), grades II–III (1 patient), grades III–IV (19 patients), and unknown grade (27 patients) [12,17–22].

Coagulation function is an important indicator of the synthetic capacity of the liver. Two studies reported PT on admission in 88 patients with a mean of 29.4 s and 14.0 s, respectively [14,16]. PT generally worsened within the first 3–4 days [14,16,18] and generally returned to normal during the 4–7 days after ingestion [16,20]. Two studies reported a mean peak PT value of 27.7 s and 55.5 s, respectively [13,17]. Four studies reported INR with values ranging from 1.1 to 10.7 on admission [12,18,21,22]. Two studies reported mean Factor V levels on admission of 33% [20] and 78% [19], and two studies reported a median Factor V nadir of 32% [17] and 21% [18], respectively, which normalized in about 4–5 days after ingestion [18,20].

Renal function

Five studies recorded admission creatinine concentrations among 156 patients with means between 0.75 mg/dL and 1.58 mg/dL [14,16,19,21,22] (0.78 mg/dL [14,16] in early presenters and 1.50 mg/dL [19,22] in late presenters). Two studies reported peak creatinine concentrations on 59 patients with medians of 0.43 mg/dL and 1.13 mg/dL [12,17], but these included some patients undergoing extracorporeal treatments. Three studies reported three cases of renal failure with an incidence of 3% (3 out of 101 cases) [16,17,20], and one study reported five cases of acute kidney injury (AKI) with an incidence of 19% (5 out of 27 cases) [18]. The diagnostic criteria for renal failure or AKI were not provided and probably differed between studies. Patients with AKI were also reported in the study of Trakulsrichai et al. [12], but the exact number of AKI patients who also had NAC treatment was not provided.

Complications in clinical course & adverse reactions to NAC

In the clinical course of amatoxin intoxication, gastrointestinal bleeding was one of the most common complications reported in patients with poor outcomes [21] and occurred in 15 out of 71 cases (21%) as reported by two studies [12,21]. The other studies did not mention the presence of gastrointestinal bleeding. Other reported complications included lactic acidosis (5%, 4 of 20 cases) [20], tracheal infection (11%, 2 of 18 cases), and arrhythmia (22%, 4 of 18 cases) [21]. Arrhythmia occurred among both survivors and patients with poor outcomes, while tracheal infections occurred only in patients with poor outcomes [21]. The most common adverse reaction to intravenous NAC treatment was anaphylactoid reactions, which was reported in one study with an incidence of 5% (4 out of 73 cases) [10]. Two other studies reported no adverse reactions related to NAC treatment during hospitalization (0/157 and 0/77 cases, respectively) [15,16].

Discussion

Considering the possible protective effects of NAC as a free radical scavenger and anti-inflammatory agent [24–27], NAC has been used for the treatment of amatoxin poisoning for decades and is one of the most frequently used drugs for this purpose in the United States [28]. *In vitro* studies revealed that amatoxin-treated hepatocytes showed significantly higher viability and lower levels of apoptosis-markers when cultured with NAC [29,30]. Although the effect of NAC is still controversial in animal models [11,31], the mortality rate of patients given NAC was among the lowest in amatoxin poisoning patients compared to those given other monotherapies or combined therapies (ranging from 5% to 35% in a 20-year retrospective analysis) [5].

In this review, the MRLTi of amatoxin-poisoning patients treated with NAC combined with other therapies was 11% (57 of 506 cases), ranging from 2.5% [15] to 44% [21], with an MRLTe of 7.9% (40 deaths in 506 cases). These large differences between studies could be partially due to the heterogeneity of each study's population, setting, or varying availability of medical resources. Nevertheless, NAC seems to be safe and effective in both pediatric and adult patients with amatoxin poisoning [10,13,14,17].

Regarding the survival of amatoxin-poisoning patients, the MRLTi of patients with NAC treatment in this review is consistent with the results of previous studies [5,32]. In the study by Akın et al. [23], the MRLTi of the NAC treatment group (4%) was lower than the MRLTi of the control group (19%), which supports the potential benefit of NAC in amatoxin-poisoning patients. Unfortunately, a selection bias is apparent in other studies. Karvellas et al. [21] only studied patients with acute liver injury or acute liver failure and had

the highest MRLTi of 44%. Similarly, Kieslichova et al. [22] only studied patients admitted to the ICU at a liver transplant center. Their mean admission ALT (2295 IU/L) and AST (2130 IU/L) illustrates that these patients were likely quite ill before reaching this transplant center. Additionally, 8 and 10 cases of grade III–IV hepatic encephalopathy were reported in these two studies, respectively [21,22], which may also account for their relatively poor outcomes. Therefore, it is highly likely that the actual MRLTi of all patients treated with NAC is even lower than 11%.

In previous studies, the reported mortality rate of amatoxin-poisoned pediatric patients was remarkably higher than adults (over 50%) [33,34]. According to the outcomes of studies involving children and teenagers in this review [10,13,14,17], the MRLTi was 14%, which is notably lower than previous reports. Even though the number of pediatric cases with complete information was small (only seven cases), it is still possible that NAC is effective in reducing mortality in this population.

The variable availability of medical resources by region, especially intensive care services, can influence outcomes [35]. In countries with enough medical resources, patients may have better outcomes. We did a sensitivity analysis by calculating the MRLTi excluding any one study to reflect the heterogeneity of included studies. Exclusion of Locatelli et al. [15] would lead to the highest MRLTi of 15%, and exclusion of Trakulsrichai et al. [12] would lead to the lowest MRLTi of 9.6%, which may be partially explained by a relative difference in available medical resources in these study locations. The outcome of amatoxin-poisoning patients not only depends on the therapy used, but also on the interval between mushroom ingestion and admission to hospital [5,7]. The MRLTi in early presenters (6.3%) [14,16–18,23] was significantly lower than the MRLTi in late presenters (23%) [13,19,20,22]. Medical resource availability and early diagnosis are very important in treating amatoxin poisoning.

Outcome indicators in acute liver failure patients include prothrombin time, serum creatinine, lactate, bilirubin, and transaminases [36–38]. Akin et al. [23] observed that AST, ALT, and INR were lower in the NAC treatment group than in the control group, which is consistent with a lower MRLTi in the NAC group. The interval from ingestion to arrival at the extremes of these indicators was very different between surviving patients (4–5 days) and those with poor outcomes (6–7 days) [19]. In amatoxin poisoning patients treated with NAC, the highest values of transaminase and prothrombin time were attained at around 3–4 days after ingestion [13,14,16,18,20].

The best combinations for treating amatoxin poisoning remain unclear. The MRLTi of combined treatments of NAC with silibinin/silymarin, penicillin G, and extracorporeal elimination methods were 10.5%, 7.0%, and 7.4%, respectively, which were all lower than overall MRLTi (11%). Although heterogeneity in these sub-analyses prevented further analysis by treatment, it is still possible that these combined treatments (with the possible exception of silibinin/silymarin) could further improve outcomes for amatoxin-poisoning patients also being treated with NAC.

The optimal dose and duration of NAC treatment for amatoxin poisoning remains unknown. Anaphylactoid reactions to intravenous NAC treatment including rash, flushing, pruritis, bronchospasm, hypotension, chest pain, and angioedema have been reported [39]. Still, only four cases of anaphylactoid reactions occurred in just one study [10]. In two other included studies, no adverse reactions occurred [15,16], while the rest of the included studies did not mention any adverse reactions related to NAC. We believe NAC appears safe for use in amatoxin poisoning patients.

This study had several limitations. First, several studies using therapies in addition to NAC were included, thus outcomes cannot be attributed solely to NAC. Second, treatment regimens and study populations were not consistent between studies. Third, no prospective study of NAC treatment for amatoxin poisoning has been reported, so this review is only based on case reports, case series and retrospective cohort studies. Evidence strength for NAC treatment in amatoxin poisoning may be enhanced by future studies with more complete information reporting. An international registry of amatoxin poisoning patients could be of immense value for further analyses.

Conclusion

NAC treatment combined with other therapies appears to be beneficial and safe in both adults and children with amatoxin poisoning. Until further data emerge, it is reasonable to use NAC in addition to other treatments for patients with amatoxin poisoning.

Author contributions

YL devised the idea for the study; JL, YC, and YL developed and tested the data extraction form; JL and YC collected the data; XL, SY, LZ, and ZG analyzed the data; JL and YC drafted the paper; and JL, YG, JW and YL revised the paper.

Disclosure statement

The authors report no conflicts of interest.

Funding

This work is supported by the National Natural Science Foundation of China [grant number 81701893 (YG) and 81550034 (YL)], the Foreign Cultural and Educational Experts Recruitment Project of the State Bureau of Foreign Experts Affairs, and the Dongcheng District Excellent Talents Program of Beijing.

References

[1] Yilmaz I, Ermis F, Akata I, et al. A case study: what doses of amanita phalloides and amatoxins are lethal to humans? Wilderness Environ Med. 2015;26:491–496.
[2] Vetter J. Toxins of *Amanita phalloides*. Toxicon. 1998;36:13–24.
[3] Wieland T. The toxic peptides from *Amanita* mushrooms. Int J Pept Protein Res. 1983;22:257–276.

[4] Garcia J, Costa VM, Carvalho A, et al. *Amanita phalloides* poisoning: mechanisms of toxicity and treatment. Food Chem Toxicol. 2015;86:41–55.
[5] Enjalbert F, Rapior S, Nouguier-Soulé J, et al. Treatment of amatoxin poisoning: 20-year retrospective analysis. J Toxicol Clin Toxicol. 2002;40:715–757.
[6] Diaz JH. Amatoxin-containing mushroom poisonings: species, toxidromes, treatments, and outcomes. Wilderness Environ Med. 2018;29:111–118.
[7] Ye Y, Liu Z. Management of *Amanita phalloides* poisoning: a literature review and update. J Crit Care. 2018;46:17–22.
[8] Yoon E, Babar A, Choudhary M, et al. Acetaminophen-induced hepatotoxicity: a comprehensive update. J Clin Transl Hepatol. 2016;4:131–142.
[9] Pertile N, Galliani E, Vergerio A, et al. The *Amanita phalloides* syndrome. Case of a 2-year-old girl. Pediatr Med Chir. 1990;12:411–414.
[10] Locatelli C, Maccarini D, Travaglia A, et al. Prolonged high-dose n-acetylcysteine treatment of amanita phalloides and chlorinated hydrocarbon poisoning. Pharmacol Res. 1992;26:201.
[11] Schneider SM, Michelson EA, Vanscoy G. Failure of *N*-acetylcysteine to reduce alpha amanitin toxicity. J Appl Toxicol. 1992;12:141–142.
[12] Trakulsrichai S, Sriapha C, Tongpoo A, et al. Clinical characteristics and outcome of toxicity from *Amanita* mushroom poisoning. Int J Gen Med. 2017;10:395–400.
[13] Yamada EG, Mohle-Boetani J, Olson KR, et al. Mushroom poisoning due to amatoxin. Northern California, Winter 1996-1997. West J Med. 1998;169:380–384.
[14] Montanini S, Sinardi D, Pratico C, et al. Use of acetylcysteine as the life-saving antidote in *Amanita phalloides* (death cap) poisoning. Case report on 11 patients. Arzneimittelforschung. 1999;49:1044–1047.
[15] Locatelli CA, Petrolini VM, Vecchio S, et al. Outcome in 157 alpha-amanitin-poisoned patients treated with the "Pavia protocol. Toxicol Lett. 2010;196:S91.
[16] Ahishali E, Boynuegri B, Ozpolat E, et al. Approach to mushroom intoxication and treatment: can we decrease mortality? Clin Res Hepatol Gastroenterol. 2012;36:139–145.
[17] Grabhorn E, Nielsen D, Hillebrand G, et al. Successful outcome of severe *Amanita phalloides* poisoning in children. Pediatr Transplant. 2013;17:550–555.
[18] Bonacini M, Shetler K, Yu I, et al. Features of patients with severe hepatitis due to mushroom poisoning and factors associated with outcome. Clin Gastroenterol Hepatol. 2017;15:776–779.
[19] Escudie L, Francoz C, Vinel JP, et al. *Amanita phalloides* poisoning: reassessment of prognostic factors and indications for emergency liver transplantation. J Hepatol. 2007;46:466–473.
[20] Bergis D, Friedrich-Rust M, Zeuzem S, et al. Treatment of *Amanita phalloides* intoxication by fractionated plasma separation and adsorption (Prometheus(R)). J Gastrointestin Liver Dis. 2012;21:171–176.
[21] Karvellas CJ, Tillman H, Leung AA, et al.; United States Acute Liver Failure Study Group. Acute liver injury and acute liver failure from mushroom poisoning in North America. Liver Int. 2016;36:1043–1050.
[22] Kieslichova E, Frankova S, Protus M, et al. Acute liver failure due to *Amanita phalloides* poisoning: therapeutic approach and outcome. Transplant Proc. 2018;50:192–197.
[23] Akın A, Keşkek ŞÖ, Kılıç DA, et al. The effects of *N*-acetylcysteine in patients with *Amanita phalloides* intoxication. J Drug Metabol Toxicol. 2013;04:5.
[24] Magdalan J, Piotrowska A, Gomułkiewicz A, et al. Influence of commonly used clinical antidotes on antioxidant systems in human hepatocyte culture intoxicated with alpha-amanitin. Hum Exp Toxicol. 2011;30:38–43.
[25] Zheleva A, Tolekova A, Zhelev M, et al. Free radical reactions might contribute to severe alpha amanitin hepatotoxicity–a hypothesis. Med Hypotheses. 2007;69:361–367.
[26] Samuni Y, Goldstein S, Dean OM, et al. The chemistry and biological activities of *N*-acetylcysteine. Biochim Biophys Acta. 2013;1830:4117–4129.
[27] de Andrade KQ, Moura FA, dos Santos JM, et al. Oxidative stress and inflammation in hepatic diseases: therapeutic possibilities of *N*-acetylcysteine. Int J Mol Sci. 2015;16:30269–30308.
[28] De Olano J, Wang JJ, Villeneuve E, et al. Current fatality rate of suspected cyclopeptide mushroom poisoning in the United States. Clin Toxicol (Phila). 2020.
[29] Magdalan J, Ostrowska A, Piotrowska A, et al. Benzylpenicillin, acetylcysteine and silibinin as antidotes in human hepatocytes intoxicated with alpha-amanitin. Exp Toxicol Pathol. 2010;62:367–373.
[30] Magdalan J, Piotrowska A, Gomułkiewicz A, et al. Benzylpenicyllin and acetylcysteine protection from α-amanitin-induced apoptosis in human hepatocyte cultures. Exp Toxicol Pathol. 2011;63:311–315.
[31] Tong TC, Hernandez M, Richardson WH, 3rd, et al. Comparative treatment of alpha-amanitin poisoning with *N*-acetylcysteine, benzylpenicillin, cimetidine, thioctic acid, and silybin in a murine model. Ann Emerg Med. 2007;50:282–288.
[32] Poucheret P, Fons F, Dore JC, et al. Amatoxin poisoning treatment decision-making: pharmaco-therapeutic clinical strategy assessment using multidimensional multivariate statistic analysis. Toxicon. 2010;55:1338–1345.
[33] Jander S, Bischoff J, Woodcock BG. Plasmapheresis in the treatment of Amanita phalloides poisoning: II. A review and recommendations. Ther Apher. 2000;4:308–312.
[34] Jan MA, Siddiqui TS, Ahmed N, et al. Mushroom poisoning in children: clinical presentation and outcome. J Ayub Med Coll Abbottabad. 2008;20:99–101.
[35] Bouchard J, Lavergne V, Roberts DM, et al. Availability and cost of extracorporeal treatments for poisonings and other emergency indications: a worldwide survey. Nephrol Dial Transplant. 2017;32:699–706.
[36] O'Grady JG, Alexander GJ, Hayllar KM, et al. Early indicators of prognosis in fulminant hepatic failure. Gastroenterology. 1989;97:439–445.
[37] Bernal W, Donaldson N, Wyncoll D, et al. Blood lactate as an early predictor of outcome in paracetamol-induced acute liver failure: a cohort study. Lancet. 2002;359:558–563.
[38] Ganzert M, Felgenhauer N, Zilker T. Indication of liver transplantation following amatoxin intoxication. J Hepatol. 2005;42:202–209.
[39] Sandilands EA, Bateman DN. Adverse reactions associated with acetylcysteine. Clin Toxicol. 2009;47:81–88. Feb