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Editorial

Dear Readers,

We present to you the last issue of our journal for 2022. In this issue, we have published 5 research articles, 1 review and 1 case report that we think you will read with pleasure and interest. We hope that your scientific support will continue to increase in 2023. We wish that 2023 will bring health, happiness and peace to the world and our country.

We would like to thank everyone who contributed to our journal for their support and contributions.

Best Regards.

Eurasian Journal of Toxicology Editorial Board

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Original Article
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Overview of Poisoning Cases in Bursa

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Abstract

Introduction: In this study, our aim is to examine the most common causes of poisoning in our city, to contribute to the epidemiological studies in our country and to ensure the efficient use of the country's resources.

Material-Method: In our study, all patients who admitted to the emergency departments of public and private hospitals in Bursa between 01.02.2020 and 31.08.2022 due to poisoning and were reported to the statistical reporting unit of Bursa Provincial Health Directorate were retrospectively evaluated.

Results: The gender differences in the causes of poisoning was seen that food-related poisoning was more common in men (n:322-78), and drug-induced poisoning was more common in women (n:363-556) and the difference was found to be statistically significant (x2=217.06, p<0.01). When we evaluated the differences in the causes of poisoning between age groups, it was seen that the lowest intoxication age group was <55 years of age (6.3%, n:111). Corrosive poisonings in the 0-13 age group constitute 72.7% (n:109) of all age groups. Methyl alcohol poisonings are most common in the 36-55 age group. In the chi-square analysis, a statistically significant difference was found between the age groups categorized in terms of the causes of poisoning (x2=641.80, p<0.01). When we look at the outcomes of the patients included in the study, the causes of poisoning in the patients who died were carbon monoxide poisoning (CO) (n:2), drugs (n:1), ethyl alcohol (n:1), were respectively. In addition, the most common cause of poisoning requiring ICU admission was drugs (n:82). When we look at the seasonal differences in the causes of poisoning, poisoning cases with drugs are seen more than other factors in every season, while CO is seen more in winter, food releated in summer and musrooms in autumn. The causes of poisoning show a statistically significant difference in terms of seasons (x2=565.35, p<0.01).

Conclusions: Analysis of local causes in a region is very important in poisoning. In this way, measures specific to that region can be taken. In this way, antitoxin planning and manpower planning can be made more effective.

Keywords: Poisoning cause, gender, mortality, Bursa region

Introduction

Poisoning is a common cause of death and disability on a global scale, and a frequent cause of emergency room visits and hospitalizations. According to WHO 2016 data, 106,683 people worldwide died in one year due to unintentional poisoning[1]. Acute poisoning cases are increasing day by day due to changes in social life and variations in chemical substances, and it seems to occupy our agenda more[2,3]. Especially, these problems are becoming a more important public health problem in underdeveloped and developing countries due to the low level of health system and infertility in the weekly life cycle[4]. According to WHO, one person dies every forty seconds due to a suicide. The vast majority of these deaths are caused by chemical agents, and globally, 79% of all are reported from underdeveloped and developing countries. This makes suicide the most common cause of death for the population between the ages of 15-29 [5].

It is seen that the share of poisonings in emergency admissions in the world is between 0.02-0.09%[6,7]. Although the frequency of poisoning and the cause of poisoning vary according to the region, it has been reported in a limited epidemiological study in our country that the frequency of poisoning varies between 0.8% and 5% among all emergency department admissions [8,9]. In the United Kingdom, the equality of men and women in poisonings between the ages of 15-35, and the high mortality in poisoning in the elderly is remarkable[10]. In the childhood group up to the age of 6, poisoning occurs most frequently when children reach and take drugs such as paracetamol and ibuprofen that are not properly stored at home, the prevalence of poisoning in the 6-11 age group is relatively low compared to other childhood periods. comes to the fore[11]. It has been reported that 150,000 cases of poisoning are seen annually in our country, 80% of which occur in childhood[12]. Poisoning with the drugs given by the parents in the first years of life, cleaning agents between the ages of 2-3, and drugs kept in the cupboard between the ages of 3-5; In school childhood and adolescence, drug poisoning for suicidal purposes is more common[13].

The most common reasons for suicide attempts are mental illness, poverty, unmet demands, chronic illness, love failure, and emotional failures[14]. It has been reported that acute poisoning is the most common reason for admission to the emergency department in developed countries, and it is the

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second most common reason for admission to the emergency department after infectious diseases in developing countries. Worldwide, poisonings constitute 5-10% of all deaths [15].

Poisonings show a national variation according to the plant and living animal species grown according to the agent used. Epidemiology of poisoning; It is important both on behalf of emergency physicians and on behalf of those who have experienced poisoning for their changing knowledge and treatment attitudes. In today's world, where information about the etiology and epidemiology of poisoning is changing rapidly, it is important for health care providers to follow the innovations in treatments[16].Therefore, regional epidemiological data; Rational planning of national resources allocated to poisoning is important because it contributes to acceleration in procurement processes, such as the necessity of having antidotes used for certain poisonings in common places.

In this study, our aim is to examine the most common causes of poisoning in our city, to contribute to the epidemiological studies in our country and to ensure the efficient use of the country's resources.

Material and Methods

In our study, all patients who admitted to the emergency departments of public and private hospitals in Bursa between 01.02.2020 and 31.08.2022 due to poisoning and were reported to the statistical reporting unit of Bursa Provincial Health Directorate were retrospectively scanned. At the planning stage of the study, preliminary permission was obtained from the Ethics Committee of Bursa City Hospital, dated 28.09.2022 and numbered 2022-11/2 from KAEK, and from the Provincial Health Directorate with the letter dated 31.08.2022 and numbered 2061 (Barcode: 172539649). The admission dates, causes of poisoning, symptoms at admission, age and gender, and discharge status of the patients were recorded. The causes of poisoning are carbon monoxide intoxication (CO), food poisoning (Food), drugs (Medicine), fungi (Mushrooms), ethyl alcohol (Ethyl A), methyl alcohol (Methyl A.), insecticides (Insectiside), corrosive substances (Corrosive) and as others (Other), age groups are 0-13, 14-25, 26-35, 36-55 and >55 also seasonally according to admission dates, winter (Winter), spring (Spring), summer (Summer), autumn (Statistical analyzes were performed by categorizing them as Autumn).

The data of the study were analyzed using the 'The jamovi project (2022) jamovi (Version 2.3) (Computer Software). Descriptive statistics were expressed as mean ± standard deviation or median values and an interquartile range (IQR) of 25–75 %, while categorical variables were expressed as numbers and percentages (%). Kolmogorov–Smirnov test and Shapiro-Wilk test were used for the normality distribution of the data. While the significance of the difference between the groups in terms of continuous numerical variables in which parametric test statistics assumptions were provided was examined with Student's t test, the significance of the difference in terms of continuous numerical variables where parametric test statistics assumptions were not met was evaluated with the Mann-Whitney U and Kruskal-Wallis tests. Chi-square and Fisher's exact test were used to analyze whether there was a relationship between categorical variables. The variables that may be effective for mortality were evaluated using the "enter" method in logistic regression analysis. $p < 0.05$ was considered statistically significant. Results were given at 95 % confidence interval.

Results

In our study, 1856 patients who admitted to the emergency departments of Bursa province hospitals due to poisoning and were notified to Bursa Provincial Health Directorate were included. Of all patients, 53.2% (n: 987) were male, and the median age of the patients was 28 (IQR: 25-75) (22:17-39). While the most poisoning cases were seen with 26% (n: 483) in the patients aged 36-55 years, the lowest rate of poisoning was seen with 6.3% (n: 117) in the 55< age group. Considering the seasonal distributions of poisonings, it was seen that while it was most common in summer with 38% (n: 705), poisoning cases were observed with a rate of 15.3% (n: 284) in autumn. The three most common causes of poisoning were 49.5% (n:919) drugs, 21.6% (n:400) foods, and 12.3% (n:229) CO. Food poisonings increased especially in summer months and CO poisonings increased in winter months. When we look at the symptom distribution of the patients at the time of admission, 55.8% (n: 1035) presented with nausea and vomiting, while 27% (n: 501) did not have any symptoms. While 62.6% (n: 1162) of the patients were discharged from the emergency department, 31.3% (n: 581) were admitted to the service, 5.8% (n:107) were admitted to the ICU, and 0.3% (n: 6) were found to be exitus.

When we evaluated the gender differences in the causes of poisoning, it was seen that food-related poisoning was more common in men (n:322-78), and drug-induced poisoning was more common in women (n:363-556). In addition, in the Pearson Chi-square analysis we conducted, a statistically significant difference was found between men and women in terms of poisoning causes ($x2=217.06$, $p<0.01$) (Table 1).

Table 2: Outcomes of the patients according to the poisining etiology

	N	Male	Female	Test Statistic
Cause of Poisoning	**1856**	**(N = 987)**	**(N = 869)**	
CO		113	116	X2 = 217.06, P <0.01
Food		322	78	
Medicine		363	556	
Mushrooms		7	16	
Ethyl Alcohol		18	8	
Metyl Alcohol		14	1	
Insecticide		28	26	
Corrosive		95	55	
Other		27	13	

Table 2: Age distribution of poisioning causes

Cause of Poisoning	N 1856	0-13 (N=375)	14-25 (N=432)	26-35 (N=449)	36-55 (N=483)	>55 (N=117)	Test Statistic
CO		27	48	51	68	35	
Food		17	50	176	138	19	
Medicine		187	301	199	203	29	
Mushrooms		0	2	0	11	10	
Ethyl Alcohol		5	4	4	9	4	X2 = 641.80, P <0.01
Metyl Alcohol		0	1	0	13	1	
Insecticide		13	10	6	16	9	
Corrosive		109	14	9	14	4	
Other		17	2	4	11	6	

When we evaluated the differences in the causes of poisoning between age groups, it was seen that the lowest intoxication age group was 6.3% (n:111) in people >

55. Corrosive poisonings in the 0-13 age group constitute 72.7% (n:109) of all age groups. Methyl A poisonings are most common in the 36-55 age group. In the chi-square analysis, a statistically significant difference was found between the age groups categorized in terms of the causes of poisoning (x2=641.80, p<0.01) (Table 2).

When we look at the distribution of discharge from the emergency department of the patients included in the study, the causes of poisoning in the patients who died were CO (n:2), Medicine (n:1), Ethyl A (n:1), Methyl A (n:1) and Other (n:1)) form. In addition, the most common cause of poisoning requiring ICU admission was Medicine (n:82). A statistically significant difference was found in terms of the outcome status of the poisoning causes of the patients (x2=238.42, p<0.01) (Table 3).

When we look at the seasonal differences in the causes of poisoning, poisoning cases with drugs are seen more than other factors in every season, while CO is seen more in winter, Food in summer and Musrooms in autumn. The causes of poisoning show a statistically significant difference in terms of seasons (x2=565.35, p<0.01) (Table 4).

Table 3: Outcomes of the patients according to the poisining etiology.

Cause of Poisoning	N 1856	Exitus (N=6)	Hospital admissions (N=581)	Discharge (N=1162)	ICU (N=107)	Test Statistic
CO		2	47	171	9	
Food		0	57	342	1	
Medicine		1	346	490	82	
Mushrooms		0	14	9	0	
Ethyl Alcohol		1	11	12	2	X2 = 238.42, P <0.01
Metyl Alcohol		1	4	5	5	
Insecticide		0	22	29	3	
Corrosive		0	60	87	3	
Other		1	20	17	2	

Table 4: Seasonal distrubiton of poisioning causes

Cause of Poisoning	N 1856	Winter (N=400)	Spring (N=467)	Autumn (N=284)	Summer (N=705)	Test Statistic
CO		128	56	31	14	
Food		36	45	14	305	
Medicine		179	279	179	282	
Mushrooms		6	1	13	3	
Ethyl Alcohol		8	9	5	4	X2 = 565.35, P <0.01
Metyl Alcohol		13	1	1	0	
Insecticide		6	20	9	19	
Corrosive		19	43	26	62	
Other		5	13	6	16	

Discussion

The share of emergency admissions due to poisoning in all emergency admissions is 0.04% in Bursa. In previous studies in our country, this rate has been reported to be between 0.8% and 5%[8, 9]. When we look at the share of poisonings in all emergency admissions on a global basis, it is seen that it varies between 0.2% and 0.9%[17-19]. A previous study in our province reported a rate of 1.76% [20]. The fact that it is close to the averages in the world shows the strength of our sampling similar to the world. We attribute the fact that the rate of poisoning in Bursa province is below the studies conducted in Turkey, that the studies reported from Turkey are generally carried out in the hospital of that province, which is the poison center, and therefore, the number of admissions related to poisoning is relatively higher than in other hospitals.

When we look at the seasonal distribution of poisonings, it was seen that the most frequent application was in summer and in July. There are studies in the world that indicate that autumn is the most common season[21]. In a study conducted in Iran, it was reported that poisonings were most common in the first spring season[22]. It has also been reported that poisoning is high in summer [23]. The fact that the season in which poisoning is most common, which is so variable in the literature, is due to regional differences. We think that the high number of poisonings in Bursa in the summer months, because of the socio-cultural structure of the dinner organizations such as societies and weddings, and that the shelf life of food is difficult in hot months. Again, the high rate of poisoning in the autumn months in Bursa is due to the geographical severity of the southeastern region in this season; We attribute it to the excessive poisoning caused by the recoil from the chimney in those who are heated by stove etc. fuel.

When we look at the genders of those who have experienced poisoning, the rate of male 53% (n=987) and female 47% (n=869) was found in our study. In studies conducted in Turkey, it was previously reported that the ratio of females to males was higher in favor of females [20]. In studies conducted in the world, the rate in favor of women has been reported[6, 24]. The high rate of poisoning in women may be effective because they are more prone to depressive states and suicidal events. Our finding, which is different from the literature, may be due to the conservative nature of Bursa province and the deficiency in reported poisonings.

Considering the age distribution in poisonings, it is observed that the majority of them are young adults. In our study, 85.6% (n=1586) were younger than 45 years of age, which is similar to studies in both our country and the world literature[20, 24, 25]. These results; It may indicate that we are developing socioeconomically and that we are young in terms of population.

Abdominal pain and vomiting, which are gastrointestinal system irritation complaints, were found to be the most common first admission complaint with 55.7% of poisonings. It was compatible with the literature of our country and the world. In developed countries, it has been reported that the first reason for admission is abdominal pain and nausea with symptoms of gastrointestinal irritation[26-28]. We think that the reason for this similarity is similar emergency admission findings in countries where therapeutic drug intake is the most common cause of poisoning.

Considering the causes of poisoning, it was observed that most of them were poisoned by drugs, etc. therapeutic chemicals. In studies conducted in developing countries such as our country, the most common cause of drug intoxication was found to be consistent with our study [20, 24, 25, 29]. The reason for this may be that sociodemographic characteristics, family ties, and economic power of individuals are similar.

In our study, it was found that the most common cause of drug intoxication was multi-drug poisoning, and in poisonings caused by a single drug, the drug derivative was found to be the most common cause of paracetamol in childhood and antidepressants starting from adolescence. While there were other researchers who found that antidepressant derivatives cause poisoning most frequently, there were studies that found different types of drugs as analgesic and anti-inflammatory as the most common cause [30]. It is thought that the reason for this is due to the differences in the prescribed drugs and the sociocultural structure.

In our study, carbon monoxide poisoning was found to be 12.4% as one of the most common causes of poisoning. In studies conducted in the USA, it has been reported that it is the second most common among all poisonings[31]. The rate of CO poisoning in Bursa was 3.23% in a study[32]. In another study conducted in Izmir, it was reported that it took the second rank among all poisonings[33]. The fact that our rate is higher than the country rate seems to be related to the fact that the Bursa region is rich in terms of southwestern winds and other winds. 37% of our patients who presented with poisoning required a 24-hour long hospital stay, including intensive care follow-up. This rate was found to be 20.2% in a previous study involving only one hospital in our city[20]. In a study conducted in our country, the rate of discharge was reported as 54%[34]. The high rate of our application may be due to the high number of applicants. Our finding is consistent with the literature, with methyl alcohol, insecticide intoxications and therapeutic drug intakes taking the first place proportionally in poisonings requiring hospitalization[35, 36].

The mortality rate in our study was 0.32%. In another study conducted in our city, it was found to be 2.5%[20]. In the world, mortality rates ranging from 0.3% to 16% have been reported[21,37]. The low rate of our study may be due to the fact that the previous study was conducted in a third-level university hospital, which received a lot of referrals.

When we look at the causes of poisoning in the patients who died, it was seen that methanol poisoning was the most common and the highest proportional reason.

Conclusion

While poisoning constitutes an important part of emergency medicine practice, it is also an important public health problem. Analysis of the regional spread and distribution of poisoning cases enables the preparation of emergency health services and the availability of necessary medications for vital interventions such as antidotes. We are of the opinion that this study, which examines and analyzes poisoning cases in our province, will make important contributions to the country's data.

References

1. Weltgesundheits organisation, W.H. Organization, and E.C.F. Environment, WHO global air quality guidelines: particulate matter (PM2.5 and PM10), ozone, nitrogen dioxide, sulfur dioxide and carbon monoxide. Geneva: World Health Organization 2021 Licence: CC BY-NC-SA 3.0 IGO.
2. Alinejad S, Zamani N, Abdollahi M, Mehrpour O. A Narrative Review of Acute Adult Poisoning in Iran. Iran J Med Sci. 2017;42(4):327-346.
3. Desalew M, Aklilu A, Amanuel A, Addisu M, Ethiopia T. Pattern of acute adult poisoning at Tikur Anbessa specialized teaching hospital, a retrospective study, Ethiopia. Hum Exp Toxicol. 2011;30(7):523-7.
4. Kaale E. A retrospective study of poisoning at Muhimbili National Hospital in Dar-Es Salaam, Tanzania. J Public Health Front, 2013. 2(1): p. 21-26.
5. WHO. mental health. Available from: https://www.who.int/mental_health/prevention/suicide/suicideprevent/en/. Accessed September 30, 2022.
6. Hanssens Y, Deleu D, Taqi A. Etiologic and demographic characteristics of poisoning: a prospective hospital-based study in Oman. J Toxicol Clin Toxicol. 2001;39(4):371-80.
7. Viccellio, P., Handbook of medical toxicology. 1st ed. Boston: Little, Brown, and Company,1993.
8. Akkose S, Bulut M, Armagan E, Cebicci H, Fedakar R. Acute poisoning in adults in the years 1996-2001 treated in the Uludag University Hospital, Marmara Region, Turkey. Clin Toxicol (Phila). 2005;43(2):105-9.
9. Mert E, Bilgin NG. Demographical, aetiological and clinical characteristics of poisonings in Mersin, Turkey. Hum Exp Toxicol. 2006;25(4):217-23.
10. Camidge DR, Wood RJ, Bateman DN. The epidemiology of self-poisoning in the UK. Br J Clin Pharmacol. 2003;56(6):613-9.
11. Thanacoody R., Anderson M. Epidemiology of poisoning. Medicine, 2020. 48(3):153-155.
12. Kalaycı, A., et al., Causes of poisoning in childhood. Journal of New Medicine, 1994. 11(1): p. 4-7.
13. Uzel N. Zehirlenmeledanismar Neyzi O, Ertuğrul T. Pediatri 2002;25:1529-38.
14. Bertolote JM, Fleischmann A, Eddleston M, Gunnell D. Deaths from pesticide poisoning: a global response. Br J Psychiatry. 2006;189:201-3.
15. Carter G, Reith DM, Whyte IM, McPherson M. Repeated self-poisoning: increasing severity of self-harm as a predictor of subsequent suicide. Br J Psychiatry. 2005;186:253-7.
16. Lee HL, Lin HJ, Yeh SY, Chi CH, Guo HR. Etiology and outcome of patients presenting for poisoning to the emergency department in Taiwan: a prospective study. Hum Exp Toxicol. 2008;27(5):373-9.
17. Karakaya A, Vural N. Acute poisoning admissions in one of the hospitals in Ankara. Hum Toxicol. 1985;4(3):323-6.
18. Ozköse Z, Ayoglu F. Etiological and demographical characteristics of acute adult poisoning in Ankara, Turkey. Hum Exp Toxicol. 1999;18(10):614-8.
19. Pinar A, Fowler J, Bond GR. Acute poisoning in Izmir, Turkey--a pilot epidemiologic study. J Toxicol Clin Toxicol. 1993;31(4):593-601.
20. Akköse Aydın Ş, Köksal Ö, Fedakar R, Emircan Ş, Durmuş O. Poisoning Cases in Adults in the Years 1996-2004. Uludağ Üniversitesi Tıp Fakültesi Dergisi 2006; 32(1): 25-27.
21. Jalali A, Savari M, Dehdardargahi S, Azarpanah A. The pattern of poisoning in southwestern region of iran: envenoming as the major cause. Jundishapur J Nat Pharm Prod. 2012;7(3):100-5.
22. Naseri K, Klani Z, Sajadi ZS, Mehrpour O, Sadeghi M.Pattern of Inpatients Acute Poisonings in Birjand City, East of Iran. 2020.
23. Mehrpour, O., Sharifi, M., & Ebrahimi, M. (2015). Pattern of Acute Pediatric Poisonings in Birjand City, East of Iran. International Journal of Medical Toxicology and Forensic Medicine, 5(4):192-200.
24. Pach J, Polewka A, Zieba A, Kroch S, Chrostek Maj J, Mikołaszek-Boba M, et al. Próba kompleksowej oceny predyktorów ryzyka ponowienia samobójstwa u pacjentów po samobójczym zatruciu lekami, hospitalizowanych w Klinice Toksykologii CM UJ w Krakowie. Cześć II. Predyktory kliniczne [A trial for the complex risk assessment of repeated suicide predictors in patients after suicidal poisoning attempts, hospitalized in the Department of Clinical Toxicology CM UJ in Krakow. II. Clinical predictors]. Przegl Lek. 2001;58(4):330-4.
25. Hatzitolios AI, Sion ML, Eleftheriadis NP, Toulis E, Efstratiadis G, Vartzopoulos D, Ziakas AG. Parasuicidal poisoning treated in a Greek medical ward: epidemiology and clinical experience. Hum Exp Toxicol. 2001;20(12):611-7.
26. Aygin D, Açıl H, Investigation of poisoning cases aged 0-18 years admitted to the pediatric emergency unit. S. EEAH Medical Bulletin, 2014. 48(1): p. 27-33.
27. Merigian KS, Woodard M, Hedges JR, Roberts JR, Stuebing R, Rashkin MC. Prospective evaluation of gastric emptying in the self-poisoned patient. Am J Emerg Med. 1990;8(6):479-83.
28. Sümer V, Güler E, Karanfil R, et al. Evaluation of the poisoning cases who applied to the pediatrics emergency unit. Turk Arch Ped 2011; 46(3): 234-40.
29. Goksu S, Yildirim C, Kocoglu H, Tutak A, Oner U. Characteristics of acute adult poisoning in Gaziantep, Turkey. J Toxicol Clin Toxicol. 2002;40(7):833-7.
30. Watson WA, Litovitz TL, Rodgers GC Jr, Klein-Schwartz W, Youniss J, Rose SR, et al.2002 annual report of the American Association of Poison Control Centers Toxic Exposure Surveillance System. Am J Emerg Med. 2003;21(5):353-421

31. Froede SM, Byers JM 3rd, Wolfgang GH, Froede RC, Jones AM, Henry TE. An analysis of toxic deaths, 1982 to 1985, Pima County, Arizona. J Forensic Sci. 1987;32(6):1676-93

32. TÜRKMEN İNANIR N,AKGÖZ S Bursa'da otopsisi yapılan karbonmonoksit zehirlenmesine bağlı ölümler. Adli Tıp Dergisi. 2005; 19(2):20-25.

33. Duman E, Akgün SA, Öztürk P, Ertürk S, Şen F, Ege B. Evaluation of poisoning cases in autopsies performed in İzmir between 1997-2000. Annual Forensic Medicine Meetings 2001 Book (25-27 April 2001, İstanbul) 313-7.

34. Ok G. Our one-year poisoning cases. First Interdisciplinary Toxicology Congress Proceedings Abstract Book. 2005; 211.

35. Shin SD, Suh GJ, Rhee JE, Sung J, Kim J. Epidemiologic characteristics of death by poisoning in 1991-2001 in Korea. J Korean Med Sci. 2004;19(2):186-94. doi: 10.3346/jkms.2004.19.2.186

36. Lamireau T, Llanas B, Kennedy A, Fayon M, Penouil F, Favarell-Garrigues JC, Demarquez JL. Epidemiology of poisoning in children: a 7-year survey in a paediatric emergency care unit. Eur J Emerg Med. 2002;9(1):9-14.

37. Dragisic T, Dickov A, Dickov V, Mijatovic V. Drug Addiction as Risk for Suicide Attempts. Mater Sociomed. 2015;27(3):188-91.

Original Article

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Evaluation of illicit drugs in pediatric emergency patients using LC-MS/MS

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Abstract

Introduction: Illicit drug use is an ever-increasing problem all over the world also reflected in emergency services as drug-induced toxicities. There is limited data about illicit drugs that pose pediatric emergency cases in our country which of most are based on immunochemical screening test results that are prone to false positivity and negativity or insufficient for detection of some drugs.

Material-Method: We established validated liquid chromatography – tandem mass spectrometry (LC-MS/MS) methods for 39 illicit drug analytes and used them to evaluate blood and urine samples of pediatric emergency patients (n=50, mean age: 15.6 y, 72% male; %28 female) along with an easy and short sample preparation step.

Results: Acceptable method validation results by means of linearity, repeatability, accuracy, sensitivity, and selectivity were achieved. In analyzes using this validated method illicit drugs were detected in 60% of patients (of these 71.4% were male). Forty percent of patients showed mixed drugs. Amphetamine-type drugs and synthetic cathinones were the most found illegal drugs in samples (12 as a single drug and 12 as a mixed drug component).

Conclusions: This study was the first to use LC-MS/MS for the determination of 39 illicit drug analytes in pediatric emergency patients in our country. LC-MS/MS is a reliable and sensitive tool for the evaluation of drug-suspected emergency patients. In this way, illicit substance use profiles that cause urgent health problems can be determined accurately, and current findings can be shared through a national network, informing physicians and toxicologists as well as authorities who make regulations regarding illicit substance policy.

Keywords: Illicit drug, Pediatric emergency, Liquid chromatography-tandem mass spectrometry, Method development

Introduction

Drug abuse among children and adolescents causes deaths and urgent health problems by both direct overdose and indirectly drug-related diseases, accidents, violence and suicide. The 2018 National Drug Report revealed that illegal drug seizures increased 214% for heroin, 20% for cannabis, 75% for cocaine, 128% for ecstasy, 162% for methamphetamine, and 53% for synthetic cannabinoids in comparison to the previous year[1]. According to the European Drug Report it is estimated that one in four students, aged between 15-16 years, uses illegal drugs, since 2011. Generally, 7% of students report multiple illegal substances for lifetime use[2]. The European School Survey Project on Alcohol and Other Drugs (ESPAD) conducted a survey on 96043 students, with the average age of 15.8, in 35 countries and found that, alcohol and tobacco use (80% and 46%, respectively) were more frequently than cannabis but cannabis users also took alcohol (96%) and tobacco (91%) simultaneously[3]. Beside these data, the emergence of synthetic cannabinoids is an even greater question. Although they bind to the same receptors as cannabis (CB1 and CB2) they show a full agonist effect causing more serious acute health problems than cannabis[4]. Cocaine, amphetamine and ecstasy are common illegal stimulant drugs used among youths; while piperazines and synthetic cathinones were less reported in the past but became more popular in recent years. Stimulants lead to serious health consequences such as cardiovascular, neurological, mental, and infectious diseases or deaths[5-9].

According to the Drug Use Survey for the Young Population, conducted by the Turkish Drug Addiction Monitoring Centre (TUBIM), drug use frequency was found as 1.5% with the average age of 13 years10. The number of deaths attributable to direct substance abuse was 500 in 2013, rising to over 900 in 2017 in the general population, while 10% of deaths included 15-19 years old individuals. The most commonly used drugs were cannabis and synthetic cannabinoids, amphetamines (mostly ecstasy), opiates (heroin, morphine, codeine) and cocaine.

There are fairly little reports about the use of illicit drug testing in poisoned patients and those trials have a number of constraints. One of the most notable is that almost all were carried out with immunochemical test kits, which

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can only determine a limited number of substances and are well-known to have a high false-positive and false-negative ratio[11]. Mass spectrometry techniques such as gas and liquid chromatography - tandem mass spectrometry (e.g., GC-MS/MS and LC-MS/MS) have more advantages in terms of detecting definitively hundreds of toxicants/drugs by a single sample analysis with high accuracy. LC-MS/MS can establish exact superiority in an emergency by shortening the time for extraction because GC-MS requires additional derivatization steps which lengthen the time[12].

In this study, we aimed to establish validated LC-MS/MS methods for the analysis of illicit drugs and then, for the first time in our country, to evaluate the biological samples (blood and urine) of patients admitted to pediatric emergency service in order to determine the prevalent drug related toxicity cases.

Material and methods

Chemicals

All reference standard materials including opiates, amphetamines, cocaine, cannabis, synthetic cannabinoids, synthetic cathinones, and internal standards (IS) were purchased from Lipomed (Arlesheim, Switzerland) (Table 1). High purity acetonitrile, methanol and isopropyl alcohol were purchased from Merck (Darmstadt, Germany); ammonium formate, ethyl acetate, dichloromethane and beta-glucuronidase (85000i, Helix pomatia), were purchased from Sigma Aldrich (Taufkirchen, Germany); ultrapure water was produced by MP Minipure water system (MES Medical, Turkey).

Instrumental Conditions

We used ultra-high-pressure liquid chromatography combined with a tandem mass spectrometry (LC-MS/MS 8030-plus, Shimadzu, Japan) with an electrospray ionization (ESI) unit employed in positive mode. The chromatographic separation was performed using a Shim-Pack Column FCODS (150 mm x 2.0 mm, 3 µm, Shimadzu). The aqueous mobile phase consisted of 10 mM ammonium formate in water, while the organic mobile phase consisted of methanol. The column oven temperature was maintained at 40ºC, the flow rate was 0.4 mL/min and the injection volume was 10 µL. The flow rates of nebulizing and drying gas were 1.5 L/min and 10 L/min, respectively. The gradient flow program was optimized for each of three groups of drugs; common drugs (CD), synthetic cannabinoids (SCb), and synthetic cathinones (SCt), with the total analyzing time of 15, 22, and 12 min respectively (Table 2). Multiple reaction monitoring (MRM) method parameters were optimized by direct injection of standard solutions. The most abundant MRM transition was selected for quantification along with qualifier ions and the retention times (RT) were determined for schedule time of all substances (Table 3).

Table 1: Sensitivity, recovery and r2 values of analytes

Analyte	LOD (ng/mL)	LOQ (ng/mL)	Recovery (%)	r^2
Synthetic cannabinoids (SCb)				1 – 20 ng/mL
JWH-018-N-pentanoic acid	0.87	2.89	98.02	0.9985
UR-144-N-pentanoic acid	0.7	2.33	100.9	0.9994
JWH-018-N-5-OH-pentyl	1.07	3.56	100.3	0.9994
JWH-073-N-2-OH-butyl	0.9	2.99	98.93	0.9990
JWH-200	1.03	3.44	101.2	0.9994
UR-144-N-5-OH-pentyl	1.06	3.53	100.2	0.9974
AM-2201	0.71	2.36	102.3	0.9974
RCS-4	0.9	2.99	100.2	0.9989
JWH-250	1.03	3.45	102.7	0.9993
XLR-11	0.82	2.74	101.8	0.9991
JWH-073	0.86	2.86	99.28	0.9999
JWH-018	0.9	2.98	101.2	0.9997
JWH-081	0.8	2.66	102.2	0.9997
UR-144	0.7	2.32	101.2	0.9991
JWH-122	0.66	2.19	101.4	0.9991
Synthetic cathinones (SCt)				
Methedrone	0.72	2.41	94,41	0.9972
A-PVP	0.7	2.35	96,51	0.9963
Buphedrone	0.48	1.61	85.5	0.9971
Bupropion	1,8	5,98	96,96	0,9962
Mephedrone	0.8	2.68	100.1	0.997
d.l-4-EMC	0.97	3.24	95.89	0.9966
Common drugs (CD)				1 – 400 ng/mL
Amphetamine	0.61	2.03	101.2	0.992
MBDB	0.41	1.36	101.3	0.9955
MDA	0.85	2.85	99.2	0.9939
MDEA	0.72	2.4	102	0.9981
MDMA	0.71	2.37	100.8	0.9946
Methamphetamine	0.57	1.89	100.1	0.9965
Codeine-6-ß-D-glucuronide	1.11	3.71	102.4	0.9987
Norcodeine	0.93	3.11	99.2	0.9927
Codeine	0.64	2.13	98.4	0.9842
Dihydrocodeine	0.69	2.32	101.9	0.9929
Heroin	0.66	2.21	101.5	0.9914
Morphine	0.62	2.07	98	0.9932
Morphine-3-ß-D-glucuronide	0.55	1.83	99.6	0.9986
Buprenorphine	0.77	2.57	100.7	0.9918
Norbuprenorphine	0.82	2.72	100.8	0.9863
6AM	0.81	2.69	101.6	0.9802
BEC	0.59	1.97	98.8	0.9933
THC-COOH (+)	0.9	2.99	99.6	0.9962

LOD: Limit of Detection, LOQ: Limit of Quantification, EMC: 1,4-Ethylmethcathinone, 6AM: Monoacetyl morphine, BEC: Benzoylecgonine, THC-COOH: 11-nor-9-carboxy-Δ9-tetrahydrocannabinol, MBDB: N-methyl-1,3-benzodioxyl-butanamine, MDA: 3,4-methylenedioxyamphetamine, MDEA: 3,4-Methylenedioxy-N- ethylamphetamine, MDMA: 3,4-Methylenedioxy-N-methylamphetamine, A-PVP: alpha-Pyrolidinovalerophenone.
LOD and LOQ are the mean values obtained from the analysis of the lowest plasma and urine QC samples. Recovery is expressed as the mean value of both plasma and urine three level QC analysis results.

Table 2: Data obtained from patients' questionnaires

		Number (n)	Percent (%)
Drug History	Substance use/abuse (total)	36	72
	Alcohol	4	8
	Glue	4	8
	Heroin	4	8
	Ecstasy	8	16
	Cannabis	4	8
	Bonzai*	2	4
	Cocaine	2	4
	Mix drug	12	24
	Unknown substance	8	16
	No drug use	12	24
Education	High school	22	44
	Primary school or dropped	12	24
	Dropped out at unknown level	4	8
	Not studying	10	20
	Special education	2	4
Living with/at	Family	38	76
	Mother or Father	4	8
	Grandmother	2	4
	Dormitory	2	4
	Apart from family	2	4
	Homeless	2	4
Drug application way	Oral	16	32
	Inhalation / smoking	8	16
	Nasal sniffing / snorting	8	16
	Injection	4	8
Gender	Male	36	72
	Female	14	28
	Mean age (10-17y, median age: 16)	15.6 ± 1.81	
Parents	Together	34	68
	Divorced	7	14
	Separated	9	18

* synthetic cannabinoid

Table 3: Drugs in patients based on LC-MS/MS analysis

	n	%	Descriptions
Drug positivity	30	60*	73% (n=22) male, 26% (n = 8) female
Multidrug use	12	40	ecstasy + a-PVP(n = 2) amphetamine + cannabis (n = 2) ecstasy + methamphetamine + methedrone (n = 2) ecstasy + methedrone (n = 2) ecstasy + cocaine + methedrone (n = 2) amphetamine + codeine (n = 2)
Single drug use	18	60	codeine (n = 2) a-PVP (n = 4) amphetamine (n = 8) cannabis (n = 2) cocaine (n = 2)
Drugs			
Ecstasy	8	26,7	MDMA, MDA, and/or MBDB positive
Amphetamine	12	40	Methamphetamine and amphetamine positive
Methamphetamine	2	6,7	
a-PVP	6	20	
Methedrone	6	20	
Cocaine	4	13,3	BEC positive
Codeine	4	13,3	CG and NC positive
Cannabis	4	13,3	THC and/or THC-COOH positive

a-PVP: alpha-Pyrolidinovalerophenone; BEC: benzoylecgonine; CG: Codeine 6-beta-D-glucuronide; MBDB: N-methyl-1,3-benzodioxyl-butanamine; MDA: 3,4-methylenedioxyamphetamine; MDMA: 3,4-Methylenedioxy-N-methylamphetamine; NC: Norcodeine; THC: Δ9-tetrahydrocannabinol; THC-COOH: 11-nor-9-carboxy-Δ9-tetrahydrocannabinol.
*of 50 patients in total

Preparation of calibrator and control samples

Urine and blood plasma samples collected from healthy and non-drug users (n=5) were verified as blank matrixes. The stock solutions (1µg/mL) of reference standard materials and IS were prepared in methanol and were stored at -20oC. Blank blood and urine samples fortified with standard materials to obtain seven serial concentrations between 1 - 400 ng/mL for CD, 1 - 20 ng/mL for SCb and SCt were used to construct the calibration curves. Positive urine and blood quality control (QC) samples with three different concentrations of 10, 75, and 300 ng/mL (CD) and 2, 8, and 20 ng/mL (SCb and SCt) were prepared daily and freshly, separately from calibrators. Each calibrator and QC sample was fortified with appropriate IS with the final concentration of 100 ng/mL (for CD) and 40 ng/mL (for SCb and SCt). Negative and two positive (low and high concentration) urine or plasma QC samples were included before and after every batch of analysis.

Method validation studies

We used QC solutions prepared in three different concentrations for the validation of three methods according to international and national guidelines[13,14]. Linearity was defined in the concentration range 1- 400 ng/mL and 1-20 ng/mL for CD and for SCb and SCt respectively, and expressed as calibration regression coefficients (r2). Selectivity and specificity were evaluated by determining the lack of interfering peaks at the interested retention times in fortified (with standard solutions) and non-fortified plasma and urine samples. Imprecision (RSD) and accuracy (bias) were calculated for intra- and inter-day, up to 5 days with five replicates of each level with the accepting criteria below 20% for both. The recovery was evaluated from the results of QC samples compared to those of neat standards with 100% recovery. Sensitivity by means of limit of detection

(LOD) and limit of quantification (LOQ) were calculated from consecutive measurements (n=10) of the lowest QC samples. Signal-to-noise ratio of the analyte response was ≥3 for LOD and ≥10 for LOQ.

Collection of blood and urine samples

The ethical approval was obtained from Clinical Research Ethics Committee (reference number: 2015-026). 140 patients (9-18 years old) out of a total of 3270 patients admitted to the pediatric emergency service between October 2017 and March 2018 were evaluated for suspected substance use (having symptoms such as unconsciousness, clouding of consciousness, agitation, tachycardia, hypertension, hypotension, nausea and vomiting). Fifty patients (35.7%) and their relatives volunteered to participate in the study and gave written informed consent. Urine and blood plasma (obtained by centrifuging of blood at 1500 xg for 10 min) samples collected from all participants were recorded anonymously and stored at -20°C until analysis. The medical history, education, and sociodemographic information of each patient were also noted after a short questionnaire.

Sample preparation

Plasma and urine samples, fortified with IS were first mixed with acetonitrile (v/v=1/1), then centrifuged for 5 min at 14000 rpm. The supernatant (200 µL) was transferred to the auto sampler vial. For evaluating THC and synthetic cannabinoids, urine samples were also subjected to enzymatic hydrolysis by using sequentially beta-glucuronidase (0.5 mL, 30 min incubation) and dichloromethane/ethyl acetate/isopropyl alcohol (1:1:3, v/v) (2.5 mL), which then evaporated under nitrogen, dissolved in methanol (50 µL) and transferred to LC vials.

Data analysis

Microsoft Excel (2017, version 15.39) program was used for all calculations and method validation studies. Calibration coefficients (r2) and sample concentrations (by comparing the signal peak area values of analytes with the peak area values of internal standards) were defined by LC-MS/MS software (Lab Solutions Version 5.80, Shimadzu). Analyte results higher than LOQ levels were accepted as positive.

Results

We achieved appropriate method validation results for all analytes individually (Table 1, 4). LOD and LOQ were defined in the range of 0.4 – 1.8 and 1.36 – 5.98 ng/mL respectively. Recovery was estimated between 85.5 – 102.7%. The average r^2 of calibration curves was calculated above 0.98. Intra- and inter-day accuracy and precision were found within acceptable ranges, all values were below 15%. Total ion chromatograms of three different methods are presented as supplemental figures (Figure 1-3). We tested sample stability by repeated analysis of the same samples with three different concentrations within day (n=3, after every 3 hours) and between days (3 days) by keeping them on the autosampler (4°C) and found the accuracy and precision below 15%. Dilution of samples up to ten times did not affected the results significantly (bias <15%). In the case of repeated high drug results in ten-fold diluted samples, we did not recalibrate the method because these high results were already sufficient to show us the high-level drug positivity.

Table 4: Amounts (ng/mL) of drugs detected in patients' samples

Drug	Urine	Blood
Amphetamine	48.6 - 21602.8	24.3 - 36.5
MDMA	11391.8 - 61570.1	259.56 - 889
MDA	315.2 - 1302.7	87 - 91
MBDB	68.2	< LOQ
Methamphetamine	42.1	< LOQ
a-PVP	4.31 - 56.9	9.2
Methedrone	21.9 - 33.2	17.7
Cocaine (BEC)	386.7	12.4 - 26.5
THC-COOH	2.9	6.1
CG	88.6 -281.2	< LOQ
NC	20.4	< LOQ
SCb	< LOQ	< LOQ

a-PVP: alpha-Pyrolidinovalerophenone; BEC: Benzoylecgonine; CG: Codeine 6-beta-D-glucuronide; MBDB: N-methyl-1,3-benzodioxyl-butanamine; MDA: 3,4-methylenedioxyamphetamine; MDMA: 3,4-Methylenedioxy-N-methylamphetamine; NC: Norcodeine; THC-COOH: 11-nor-9-carboxy-Δ9-tetrahydrocannabinol; SCb: Synthetic cannabinoids.
Data are presented as amount ranges for those results obtained from more than one sample and as a single result for those results obtained from one sample.

Seventy-two percent (n=36, 72%) of patients stated using any substance including volatiles during their questionnaire (Table 2). LC-MS/MS analysis (volatiles were not evaluated) revealed that 60% out of 50 patients were drug positive of which 73% were male (Table 3). Forty percent of drug positive patients showed multidrug. Detected substances were amphetamines, (amphetamine, methamphetamine, ecstasy) synthetic cathinones (alpha-PVP, methedrone), cocaine, codeine and cannabis. Ecstasy (MDMA, 3,4-Methylenedioxy-N-methylamphetamine) was the drug with the highest amount in the samples (Table 4). Synthetic cannabinoids were not detected above LOQ levels.

All patients brought to the emergency department with suspected illegal drug misuse were observed in accordance with the advice of the National Poison Control Centre. They were monitored and vital parameters were followed at frequent interval. None of them required any indication of intervention or intensive care follow-up. All patients were discharged from the emergency room after the normalization of their vital signs and test results. Patients with their relatives were directed to the Department of Social Work and the Department of Child and Adolescent Psychiatry for consultation.

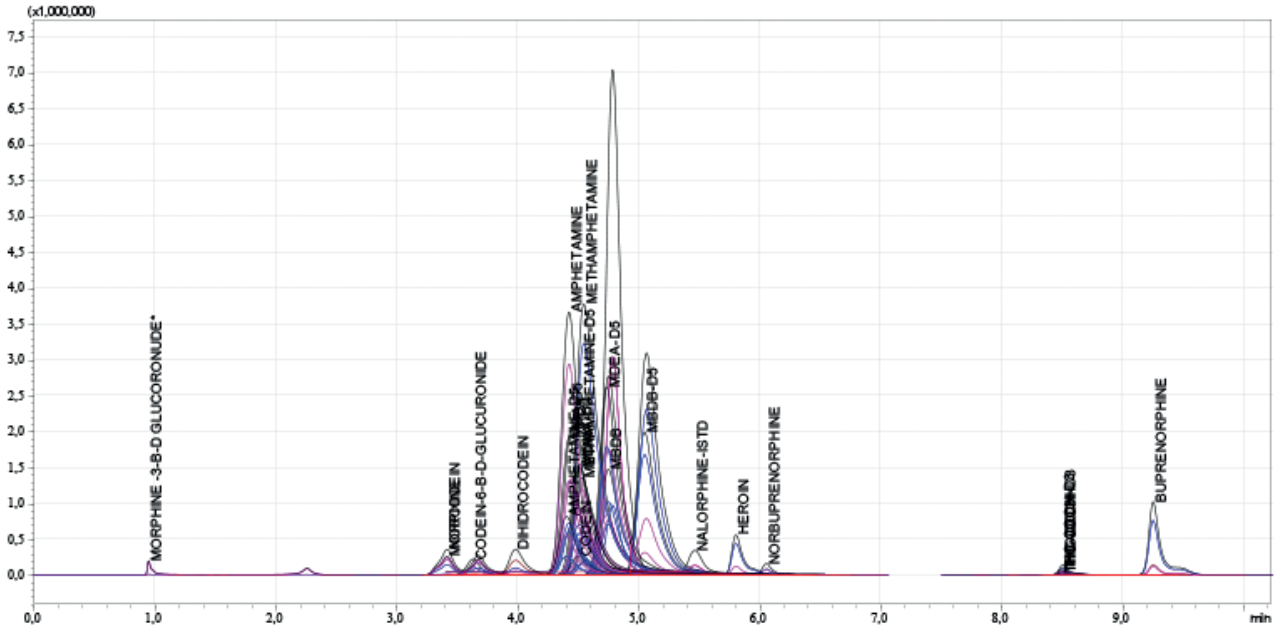

Figure 1: Total ion chromatogram of common drugs

Discussion

Regarding our method development and validation studies, similar results are also reported in literature[15-21]. We did not perform freeze and thaw studies in our validation experiments. We held our patients' samples maximum for 6 weeks at -20°C until analysis, control samples for 4 weeks at 4°C, and all samples for 6 hours at 4°C (in the autosampler) during LC-MS/MS analysis. Neither of these conditions appears to cause a significant change in the stability of the analytes according to published articles (16, 22-26).

The previous survey data on drug use preferences in adolescents indicated that amphetamines were less frequently

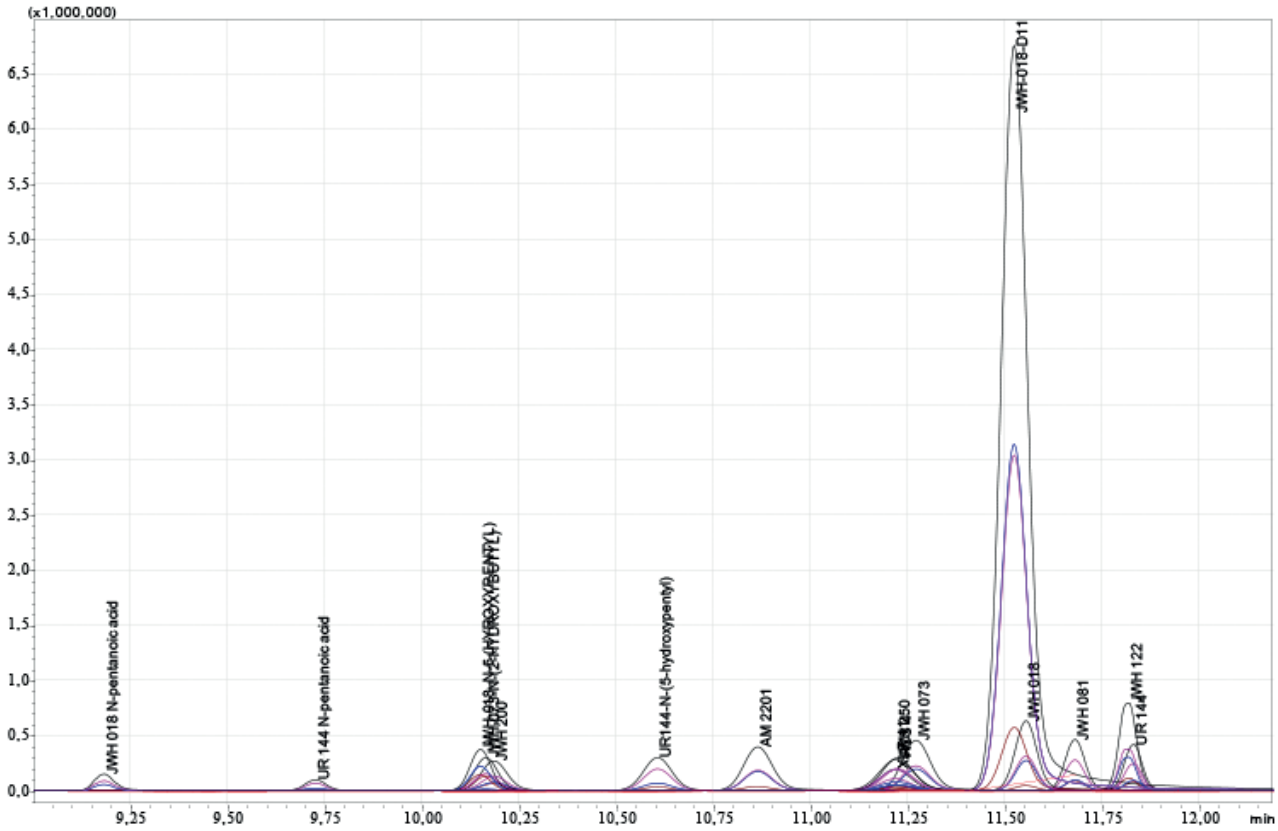

Figure 2: Total ion chromatogram of synthetic cannabinoids

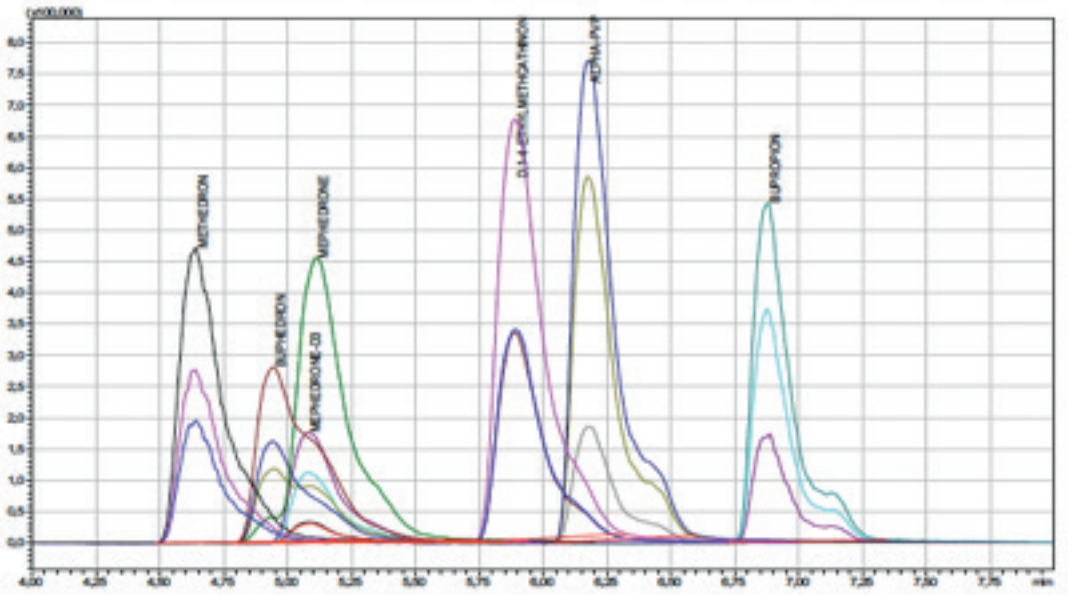

Figure 3: Total ion chromatogram of synthetic cathinones

preferred substances (22.7%) than cannabis/cannabis derivatives (84.1%) and volatiles (32.9%)1. Cannabis is still among the most widely misused drugs both in youth and adults. However, synthetic designer drugs cause more acute health problems due to their potent side effects. In addition, the production and variety of these synthetics are constantly increasing. The number of ecstasy seizures in Europe rose up from 3 million tablets in 2010 to over 15 million tablets in 2017, and our country attracts the attention where MDMA was seized more than double the amount in the last year (8.6 million in 2016)[10], which could explain why amphetamines were frequently detected in patients' samples.

In patients with heroin use history we found codeine metabolites, which indicates that they might have been taken codeine or heroin, since illegal heroin (also called diacetylmorphine, is derived from the opium alkaloid morphine, where other alkaloids such as codeine, thebaine and papaverine are also ingredients of opium) is mainly found mixed with codeine. Besides codeine is a natural alkaloid found in opium poppy[27] (legally consumed as food, but opium alkaloids are regarded to be reduced during food processing), it is also a medicine used in pain treatment that can be abused. In these cases, heroin (involving codeine) might have taken days ago, that's why we could not detect its metabolites such as 6AM (6-Monoacetyhlmorphine) (specific for heroin) and morphine, but we detected codeine in urine appeared as codeine-6-glucuronide and norcodeine, confirming that the body was exposed to codeine recently and metabolized it. In two of heroin use-stated patients, we also found amphetamine. Amphetamine might have been detected due to the use of illegal amphetamine or other drugs that were metabolized to amphetamine; but, none of the patients stated an intake of such drugs (e.g. selegiline, benzphetamine, chlorobenzorex, dimethylamphetamine, ethylamphetamine etc). Two patients with the statement of stone (crack cocaine) use have been confirmed with the results of analysis both in urine and blood. We detected multidrug in 12 patients (40%) as told by the same number of patients initially, where amphetamines were combined with cathinones, cocaine, codeine, and cannabis (Table 3). With these findings we determined the real drug type(s) taken or abused by the patients by using validated LC-MS/MS methods. Most of these drugs such as synthetic cathinones could not be detected if immunochemical drug screening would have been applied, because there are no test kits for these analytes.

Looking at reports from different countries; in a prospective cohort of Israeli adolescents (n: 138, median age: 16 years, gender: 47% male) admitted to emergency department 28% had a history of substance abuse, but the laboratory results showed a positivity of 5% for THC, 4% for opioids, 4% for MDMA (revealed after immunochemical urine drug screen test), and 29% for ethanol (in blood)[28]. Among patients presented to US pediatric emergencies between 1997-2010, 58.8% was found positive for illicit drugs, from which 4.5% was opioids, 5% stimulants (cocaine, amphetamine etc.), 6% cannabis, and 43.5% other (unspecified or combined), indicating that stimulants were most frequent drugs as found in our current study[29]. Among 30 drug positive patients we found 73.4% positivity for amphetamines, 40% for synthetic cathinones, and 13.3% for cocaine, codeine, and cannabis, and many of these drugs were used in combination (40%).

Showing the presence of synthetic cathinones (a-PVP and methedrone) in our patients' samples was interesting because there were almost no confirmed antemortem data about cathinone use in our country among children or adolescents. Just as in synthetic cannabinoids, cathinones

are also being produced with new chemical formulas and novel drugs are being marketed under the name of plant, incense and bath salt as legal substances. These synthetic drugs can attract the attention of children because of their cheapness and especially easy availability on the internet. As reported in an article, 51 patients under 20 years of age were admitted to the Texas Poison Center between the years 2010 and 2011 with synthetic cathinone exposure which of 60.8% were male with a mean age of 17.5 (age range 12-19 years), 74% had serious health problems30. The European Early Warning System (EWS) Report revealed that 60% of the substances reported in 2014 were new psychoactive substances (NPS) (especially synthetic cannabinoids and cathinones), but much reduced and remained similar in the years 2017 and 2018 (55 NPS)[10]. In two of our patients having a history of bonzai use, we detected alpha-PVP and amphetamine in their urine samples respectively, confirming the knowledge that bonzai besides synthetic cannabinoids also may involve other synthetic designer drugs such as cathinones, and amphetamines. None of our patients showed a positive result (most were between LOD and LOQ levels) for synthetic cannabinoids. It is possible to say that synthetic cannabinoids are no more preferred because of their serious acute and fatal side effects experienced by users during the last years; or less likely, newly produced synthetics, not included in our test panel, might have been missed. But confirming our findings, the number of hospitalizations in pediatric emergency departments due to using NPS decreased by more than a half in 2018 compared to 2016[31].

The fact that amphetamine-type drugs were predominantly detected in our patients' samples, suggests that adolescents may have used these substances for purposes such as keeping fit, making their minds open, and facilitating learning because they may be worried or stressed about the university exam held at the end of the high school. More importantly, pubertal changes, entertainment (for getting high), social and family problems should not be ignored as they may lead the teens to experience illegal drugs, unfortunately.

The limitation of this study may be that we did not include therapeutically used (legal) but abused drugs such as benzodiazepines, barbiturates, synthetic opioids, and other some newly produced NPS in our research panel, mainly because of our limited budget. We intended to evaluate illegal drugs that were currently seized by the national police and were widely abused, and are more prone to cause emergent health problems.

Conclusion

We developed and validated LC-MS/MS methods for the determination of multiple illegal drugs simultaneously in both blood and urine human samples with easy and short sample preparation and analysis time which have been applicated to drug suspected pediatric emergency cases for the first time in our country. It would take approximately one hour (except hydrolysis) to report the results when using these methods in an emergency laboratory, not much longer than an immunochemical drug screening test. Our findings emphasized that stimulant synthetic drugs such as ecstasy, amphetamine, alpha-PVP (colloquially called "flakka"), methedrone, and cocaine (crack) in turn were currently the most drugs related to urgent health problems, which should be taken into account by emergency physicians and toxicologists. To control and follow up the drug use, repeated studies should be performed by applying sensitive laboratory techniques, and also important preventive measures (against abused drugs and drug trafficking) should be provided by the authorities.

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References

1. EMCDDA (European Monitoring Centre for Drugs and Drug Addiction). European Drug Report 2018. 2018.Access date: 05.12.2021 http://www.narkotik.pol.tr/kurumlar/narkotik.pol.tr/Arsiv/TUBIM/Documents/Avrupa%20Uyu%C5%9Fturucu%20Raporu%202018%20EN.pdf
2. EMCDDA (European Monitoring Centre for Drugs and Drug Addiction). European drug report 2015. 2015.Access date: 06.12.2021 https://www.emcdda.europa.eu/system/files/publications/974/TDAT15001ENN.pdf
3. ESPAD (European School Survey project on Alcohol and Drugs) Report 2015 - Results from the European School Survey Project on Alcohol and Other Drugs. 2016:99.Access date: 25.12.2021 https://op.europa.eu/en/publication-detail/-/publication/2373452c-7975-11e6-b076-01aa75ed71a1/language-en
4. Tait RJ, Caldicott D, Mountain D, Hill SL, Lenton S. A systematic review of adverse events arising from the use of synthetic cannabinoids and their associated treatment. Clin Toxicol (Phila). 2016;54(1):1-13.
5. Richards JR, Garber D, Laurin EG, Albertson TE, Derlet RW, Amsterdam EA, et al. Treatment of cocaine cardiovascular toxicity: a systematic review. Clin Toxicol (Phila). 2016;54(5):345-64.
6. Swepson C, Ranjan A, Balasubramaniam M, Pandhare J, Dash C. Cocaine Enhances HIV-1 Transcription in Macrophages by Inducing p38 MAPK Phosphorylation. Front Microbiol. 2016;7:823.

7. Paratz ED, Cunningham NJ, MacIsaac AI. The Cardiac Complications of Methamphetamines. Heart Lung Circ. 2016;25(4):325-32.
8. Sanchez-Ramos J. Neurologic Complications of Psychomotor Stimulant Abuse. Int Rev Neurobiol. 2015;120:131-60.
9. Shelton CP, Rosini JM. Multisystem Organ Failure and Death Resulting From Ingestion of "Molly" (3,4-Methylenedioxymethamphetamine). J Emerg Nurs. 2015;41(5):447-50.
10. EMCDDA. European Drug Report 2019. 2019.
11. Saitman A, Park H-D, Fitzgerald RL. False-positive interferences of common urine drug screen immunoassays: a review. Journal of analytical toxicology. 2014;38(7):387-96.
12. Van Wijk XM, Goodnough R, Colby JM. Mass spectrometry in emergency toxicology: Current state and future applications. Critical reviews in clinical laboratory sciences. 2019;56(4):225-38.
13. Scientific Working Group for Forensic Toxicology (SWGTOX), Standard practices for method validation in forensic toxicology. Journal of Analytical Toxicology. 2013;37(7):452-74.
14. Ministry, of, Health. The Working Procedures and Principles of Confirmation Laboratories. Official Gazette of Republic of Turkey. 2015(Circular 14).
15. Knittel JL, Holler JM, Chmiel JD, Vorce SP, Magluilo J, Jr., Levine B, et al. Analysis of Parent Synthetic Cannabinoids in Blood and Urinary Metabolites by Liquid Chromatography Tandem Mass Spectrometry. J Anal Toxicol. 2016;40(3):173-86.
16. Jang M, Shin I, Kim J, Yang W. Simultaneous quantification of 37 synthetic cannabinoid metabolites in human urine by liquid chromatography-tandem mass spectrometry. Forensic Toxicology. 2015;33(2):221-34.
17. Marinetti LJ, Antonides HM. Analysis of synthetic cathinones commonly found in bath salts in human performance and postmortem toxicology: method development, drug distribution and interpretation of results. J Anal Toxicol. 2013;37(3):135-46.
18. Yang HS, Wu AH, Lynch KL. Development and Validation of a Novel LC-MS/MS Opioid Confirmation Assay: Evaluation of beta-glucuronidase Enzymes and Sample Cleanup Methods. J Anal Toxicol. 2016;40(5):323-9.
19. Schaefer N, Peters B, Schmidt P, Ewald AH. Development and validation of two LC-MS/MS methods for the detection and quantification of amphetamines, designer amphetamines, benzoylecgonine, benzodiazepines, opiates, and opioids in urine using turbulent flow chromatography. Analytical and bioanalytical chemistry. 2013;405(1):247-58.
20. Berg T, Lundanes E, Christophersen AS, Strand DH. Determination of opiates and cocaine in urine by high pH mobile phase reversed phase UPLC–MS/MS. Journal of Chromatography B. 2009;877(4):421-32.
21. Scheidweiler KB, Desrosiers NA, Huestis MA. Simultaneous quantification of free and glucuronidated cannabinoids in human urine by liquid chromatography tandem mass spectrometry. Clinica chimica acta. 2012;413(23-24):1839-47.
22. Ammann D, McLaren JM, Gerostamoulos D, Beyer J. Detection and Quantification of New Designer Drugs in Human Blood: Part 2 Designer Cathinones. Journal of Analytical Toxicology. 2012;36(6):381-9.
23. Wohlfarth A, Scheidweiler KB, Chen XH, Liu HF, Huestis MA. Qualitative Confirmation of 9 Synthetic Cannabinoids and 20 Metabolites in Human Urine Using LC-MS/MS and Library Search. Anal Chem. 2013;85(7):3730-8.
24. Coles R, Kushnir MM, Nelson GJ, McMillin GA, Urry FM. Simultaneous determination of codeine, morphine, hydrocodone, hydromorphone, oxycodone, and 6-acetylmorphine in urine, serum, plasma, whole blood, and meconium by LC-MS-MS. Journal of Analytical Toxicology. 2007;31(1):1-14.
25. Jimenez C, De la Torre R, Ventura M, Segura J, Ventura R. Stability studies of amphetamine and ephedrine derivatives in urine. Journal of Chromatography B. 2006;843(1):84-93.
26. Clauwaert KM, Van Bocxlaer JF, De Leenheer AP. Stability study of the designer drugs "MDA, MDMA and MDEA" in water, serum, whole blood, and urine under various storage temperatures. Forensic science international. 2001;124(1):36-42.
27. Ozbunar E, Aydogdu M, Doger R, Bostanci HI, Koruyucu M, Akgur SA. Morphine Concentrations in Human Urine Following Poppy Seed Paste Consumption. Forensic Sci Int. 2019;295:121-7.
28. Kozer E, Bar-Hamburger R, Rosenfeld NY, Zdanovitch I, Bulkowstein M, Berkovitch M. Illicit drug and alcohol users admitted to the pediatric emergency department. Age (yrs, median and range). 2008;16:12-8.
29. Fahimi J, Aurrecoechea A, Anderson E, Herring A, Alter H. Substance abuse and mental health visits among adolescents presenting to US emergency departments. Pediatric emergency care. 2015;31(5):331.
30. Forrester MB. Adolescent synthetic cathinone exposures reported to Texas poison centers. Pediatric emergency care. 2013;29(2):151-5.
31. Jalali R, Dmochowska P, Godlewska I, Balmas J, Młynarska K, Narkun K, et al. Designers Drugs—A New Challenge to Emergency Departments—An Observational Study in Poland. Medicina. 2020;56(7):354.

Original Article

Eurasian Journal of Toxicology

Protective Effects of Chlorella Vulgaris in Alcohol Intoxication

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Abstract

Objectives: The aim of the study, to investigate the effect of *Chlorella vulgaris* on the liver, kidney and heart MAPK (Mitogen-activated protein kinase), lipid peroxidation antioxidant enzyme activity with ethyl alcohol toxification.

Materials and Methods: 10-12 monthly, weighing 200-250 gr, 24 adult male Sprague Dawley rats were used. Rats were divided into 3 (n=8) groups which 2 experiments and a control. 5mg/kg of isocaloric maltose was given to the control group by gavage. 15 g/kg ethyl alcohol diluted with 50% water was given to the alcohol group and 300 mg/kg *C. vulgaris* and then 15 g/kg ethyl alcohol diluted with 50% water were given to *C. vulgaris* group. At the end of the experiment tissue samples were taken. Blood samples were collected into EDTA tubes and the tissues were kept at -20°C. The blood and tissue samples were used to investigate the GSH/GSH-Px, MAPK activity and MDA levels.

Results: MAPK activities in liver and lung tissue were increased with *C. vulgaris* which decrease with ethyl alcohol while MAPK activities in kidney and heart tissue decreased with *C. vulgaris*. The reduction in tissue GSH-Px levels with alcohol was increased significantly with *C. vulgaris* application ($p<0.05$). The declining GSH levels of liver and kidney tissue with alcohol were found to significantly increase with *C. vulgaris* ($p<0.05$). When tissue MDA levels of groups were compared, increasing MDA levels with alcohol in liver and heart tissues were determined to significantly decrease with *C. vulgaris* ($p<0.001$, $p<0.05$ respectively).

Conclusions: As a conclusion, *C. vulgaris* increased antioxidant enzyme activity especially in liver tissues and decreased lipid peroxidation in tissue which arises with ethyl alcohol that was evaluated this effect has tissue protective activity of *C. vulgaris*. Also, increasing in liver tissue MAPK activities as a result of alcohol toxication was fixed with *C. vulgaris*. This situation may be associated with a protective effect on the intracellular enzyme system activity of *C. vulgaris*.

Keywords: Ethyl alcohol, Chlorella vulgaris, GSH, GSH-Px, MAPK, MDA.

Introduction

Ethyl alcohol, which is used in the production of alcoholic beverages, is easily absorbed into the blood from all parts of the digestive system. The level of alcohol in the blood usually reaches its highest level in 45-60 minutes, depending on the fullness of the stomach and the rate of alcohol intake[1]. Behavioral disorders, manifested by decreased reflexes and muscle dissonance, occur due to toxication due to excessive alcohol consumption. Depending on the severity of the toxicity, liver enzyme dysfunction and death due to respiratory failure occur[2,3]. 90% of alcohol is metabolized in the liver and 10% in the lungs at 15 mg/dl per hour. Alcohol creates oxidative stress in tissues and affects a wide variety of cellular targets rather than a specific tissue area[4,5,6], causing many adverse metabolic disorders, especially liver damage[7,8]. About 20% of people diagnosed with alcoholism develop irreversible liver damage and severe liver disease[9,10,11].

As a result of the deterioration of the oxidative balance, "oxidative stress" occurs. Oxidative stress resulting in cellular destruction is accepted as an indicator of the deterioration of metabolic balance. The main elements that play a role in the detoxification mechanisms required by metabolism to prevent cell and tissue damage due to oxidative stress are enzymes, proteins, vitamins, plant polyphenols and derivatives that are activated to reduce or eliminate the damage caused by oxidant molecules[12,13]. Alcohol causes oxidative stress and therefore many metabolic problems[14,15]. Oxidative stress damages lipid, protein and DNA cell components, causing cell destruction and death. Superoxide dismutase, catalase and glutathione peroxidase play an active role in cell repair mechanisms[16,17]. Free radicals cause lipid peroxidation by the effect of oxidizing agents on fatty acids in the structure of membranes and lipoproteins. Malondialdehyde (MDA), a reactive aldehyde derivative, is formed as a result of lipid peroxidation caused by free radicals. The amino groups of MDA proteins react with phospholipids or nucleic acids to damage membrane structures and cause cellular damage. Changes in cellular damage are also an indicator of oxidative damage[18,19].

Antioxidants can be endogenous or exogenous. Superoxide dismutase (SOD), glutathione (GSH), glutathione peroxidase (GSH-Px) and catalase are antioxidants in enzyme structure. GSH helps transport amino acids across cell membranes, and the GSH sulfhydryl

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group is used to reduce peroxides formed during oxygen transport. In addition, GSH acts as a very important defense mechanism against peroxides formed as a result of ethyl alcohol metabolism reactions. Oxidative stress is effective in the progression of alcoholic liver disease, and antioxidants fight this stress. Antioxidant mechanisms such as GSH and GSH-Px have high activity to prevent damage[20,21].

Chlorella is the most important member of the *Chlorellaceae* family from the *Chlorophyta* group. *C. vulgaris* is rich in vitamins, proteins, minerals, amino acids, nucleic acids, essential fatty acids, enzymes and carotenoids, alpha-beta carotene, alpha-tocopherol, lycopene, lutein and zeaxanthin. Due to its rich content of *Chlorella vulgaris*, it has become a popular food supplement in oxidative stress studies in terms of preventing cell damage and death[22,23].

Mitogen-activated protein kinase (MAPK) is the familiar name for a set of receptor-mediated proteins that play a vital role in signal transduction in all eukaryotic cells from yeast to humans. It plays an important role in transporting the information sent from the receptors to the cell to the nucleus. MAPKs are proteins with apoptotic function, responsible for signal transduction and involved in the regulation of gene expressions and cell functions[24]. Alcohol use is associated with liver disease, neurotoxicity, hypertension, cardiomyopathy, immune response patterns, and increased cancer risk. Available data suggest that the MAPK family plays a central role in these alcohol-affected processes. The role of MAPK may differ depending on the cell type and the type of chronic or acute administration. For example; Acute alcohol administration causes increased MAPK activation in hepatocytes, astrocytes, and vascular smooth muscle cells. In the studies carried out, the effect processes of alcohol in such pathological conditions are not fully understood[37].

Materials and Methods

The study was carried out by the principles of Kafkas University KAU Animal Experiments Local Ethics Committee (KAU-HADYEK Decision No: 2014-31). In this study, 24 adult male Sprague Dawley rats, 10-12 months old, weighing 200-250 g, were used. The rats were kept at 25°C under standard light and 12 hours a day/12 hours in the dark and were fed ad libitum with water and food.

Experimental Groups

Rats were divided into 3 groups and each group consisted of 8 animals, 2 experimental and 1 control group. The control group was given 5 mg/kg of isocaloric maltose every 12 hours by gavage. Ethyl alcohol was given to the rats in the alcohol group (n=8) at 15g/kg/day and diluted with 50% water, and 300 mg/kg of *Chlorella* and then 50% was given to the rats in the *Chlorella* group (n=8). It was diluted with water and given 15g/kg of ethyl alcohol. The study was repeated for 20 days.

The groups used in the study were formed as follows.

Control Group (n=8, male): 5 mg/kg isocaloric maltose by gavage

Alcohol Group (n=8, male): 15 g/kg/day Ethyl alcohol + 50% water

Chlorella Group (n=8, male): First 300 mg/kg *Chlorella*, then 15 g/kg/day Ethyl alcohol + 50% water

At the end of the experiment, the anterior abdominal wall of the rats was opened with an incision under ether anesthesia. The rats, whose blood was taken by puncture by reaching the heart from the diaphragm, were sacrificed. Blood samples taken intracardially into EDTA tubes were centrifuged at + 4°C, 3000 rpm for 5 minutes and plasmas and homogenates prepared by the methods were placed in polyethylene tubes and stored at -20°C until laboratory procedures. The blood samples taken were used to measure MDA, MAPK, GSH and GSH-Px values.

Biochemical Analysis

MAPK levels were determined using the RAT MAPK14 (Mitogen-activating protein kinase 14) ELISA Kit (Elabscience Biotechnology Co. Ltd., P.R.C).

Malondialdehyde (MDA), a secondary product of lipid peroxidation (LPO), is formed by incubation of plasma with thiobarbituric acid (TBA) at 100°C. The resulting MDA forms a pink complex with TBA. By measuring the pink color at 532 nm in the spectrophotometer, LPO is determined as nmol/ml[25]. 1,1,3,3 tetra ethoxy propane was used for drawing the standard curve.

The measurement of the thiol group by enzymatic or chemical processes by dissolving the sulfhydryl group of GSH in acid forms the basis of the quantification of this compound. The absorbance values determined at 412 nm are measured as μmol/ml[26].

Glutathione peroxidase activity was measured spectrophotometrically at 412 nm in Ellman's reagent, which is based on the principle of GSH reduction in enzymatic reactions, in which cumene hydroperoxide and reduced GSH are used as co-substrates[27].

Statistical Analysis

In statistical calculations, the ONE-WAY ANOVA test was used to compare the changes in the experimental groups compared to the control groups. Results were determined as mean ± standard deviation (X ± SD) and $p<0.05$ showed the statistical difference. All calculations were made using the SPSS (16.0) package program.

Results

Statistical analysis of the experimental work of all groups is shown in the figures.

***Changes Determined in MAPK Levels of Kidney, Heart and Liver Tissues of the* Groups**

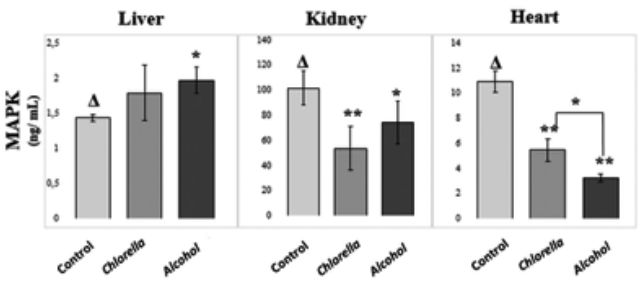

Figure 1: MAPK Levels ($^{\Delta}$* = p<0.05, $^{\Delta}$** = p<0.001)

It was determined that the values of the experimental groups were statistically significantly higher than the control group (p<0.05).

Changes in liver MAPK levels when compared between the control group and the alcohol group, it was determined that the liver MAPK levels were statistically significantly higher in the alcohol group (p<0.05). In the comparison made between the control group and the *Chlorella* group, it was determined that the liver MAPK levels did not show a statistical difference.

In the comparison between the control group and the *Chlorella* and Alcohol groups, it was determined that the kidney MAPK levels were statistically lower in the *Chlorella* and Alcohol groups (Respectively p<0.001, p<0.05).

In the statistical comparison between the control group and the *Chlorella* and Alcohol groups, a significant decrease was found in the Cardiac MAPK levels of the *Chlorella* and Alcohol groups (p<0.001). When *Chlorella* and Alcohol groups were compared, it was determined that there was a statistically significant decrease in the Alcohol group compared to the *Chlorella* group (p<0.001).

Changes Determined in MDA Levels of Kidney, Heart and Liver Tissues of the Groups

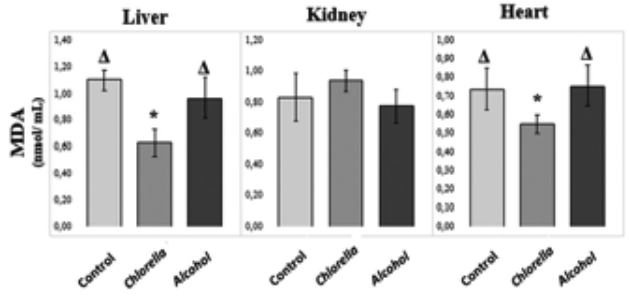

Figure 2: MDA Levels ($^{\Delta}$* = p<0.001)

A statistically significant difference was found between the Control and Alcohol groups and the *Chlorella* groups in liver MDA levels (p<0.001). It was determined that the MDA levels of the *Chlorella* group were statistically lower than the alcohol group (p<0.001).

There was no statistically significant difference in kidney tissue MDA levels.

A statistically significant difference was found between the Control and Alcohol groups and the *Chlorella* groups in the heart MDA levels (p<0.05). It was determined that the heart MDA levels of the *Chlorella* group were statistically lower than the alcohol group (p<0.05).

Changes Determined in GSH Levels of Kidney, Heart and Liver Tissues of the Groups

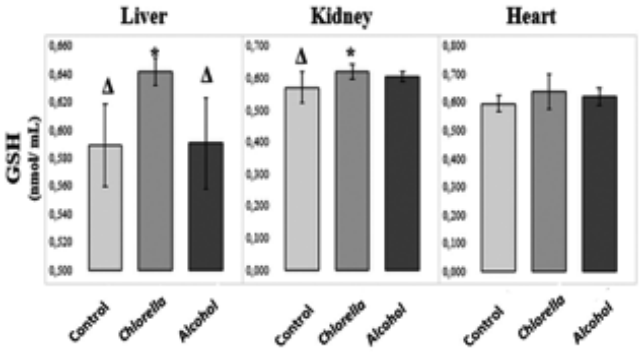

Figure 3: GSH Levels ($^{\Delta}$* = p<0.05)

When examined in terms of GSH levels in the liver tissue, it was determined that there was a statistically significant increase in the *Chlorella* Group compared to the Control and Alcohol groups (p<0.05).

When the kidney tissue GSH levels were compared, a statistically significant increase was found in the GSH enzyme levels in the *Chlorella* group compared to the control group (p<0.05).

No statistically significant difference was found when the changes in heart tissue GSH levels were compared.

Changes Determined in GSH-Px Levels of Kidney, Heart and Liver Tissues of the Groups

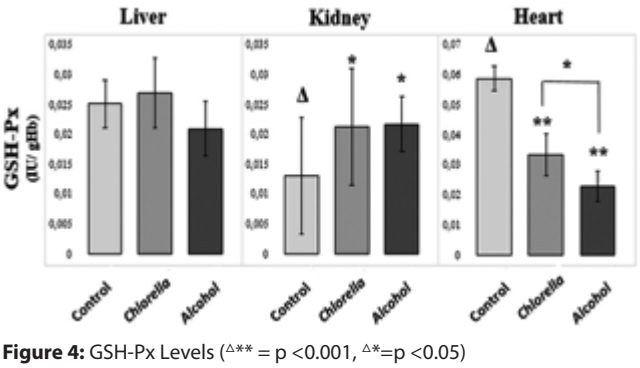

Figure 4: GSH-Px Levels ($^{\Delta}$** = p <0.001, $^{\Delta}$*=p <0.05)

In the comparison between the groups, it was determined that the changes in liver GSH-Px levels did not show a statistically significant difference.

When the changes in kidney GSH-Px levels were compared, no statistically significant difference was observed between the *Chlorella* and Alcohol groups. In the comparison between the kidney GSH-Px levels of the control group and the *Chlorella* and Alcohol groups, it was found that the GSH-Px levels of the *Chlorella* and Alcohol groups were statistically high (p<0.05).

In the comparison between the control group and the *Chlorella* and Alcohol groups, it was determined that the heart GSH-Px levels were statistically significantly lower in the *Chlorella* and Alcohol groups ($p<0.001$). In the comparison between the *Chlorella* group and the Alcohol group, it was found that the heart GSH-Px values were statistically higher in the *Chlorella* group than in the Alcohol group ($p<0.05$).

Discussion

Alcohol toxicity is caused by other metabolic products of ethanol, especially reactive oxygen species (ROS) produced during the biotransformation of ethanol. Alcohol is a strong oxidant; the Mitochondrial respiratory chain activates intracellular ROS production pathways including xanthine oxidase and nicotinamide adenine dinucleotide phosphate (NADPH) oxidase pathways. As a result, there is an increase in the production of superoxide (O_2-•), hydrogen peroxide (H_2O_2) and hydroxyl (•OH) radicals in the cell. High ethanol consumption reduces intracellular antioxidant capacity. In the damage caused by alcohol, antioxidant systems protect hepatocytes and are activated by supporting the cell membrane against the increase of radicals, and the imbalance in the cell results in the oxidation of protein, lipid and DNA. In rats, MDA, hydroperoxide and conjugated diene levels were increased due to alcohol. However, ascorbic acid application prevented the increase in free radical levels, and GSH and GSH-Px levels that decreased as a result of ethyl alcohol applications increased again after ascorbic acid application28. The P-450 enzyme system, which plays an active role in alcohol metabolism, causes ROS production in hepatocytes and causes damage[29]. As a result of the increase in the alcohol level in the blood, the cytochrome C level decreased and accordingly, it caused the formation of superoxide radicals in the liver[30]. Toxic doses of ethyl alcohol combine with superoxides in liver cells to initiate lipid peroxidation and the ROS products formed trigger cellular deterioration. DNA breaks caused by free radicals can be prevented by applying metallothionein, which is a rich protein source[31].

In our study, we determined that alcohol application increased lipid peroxidation in liver and heart tissue, while *Chlorella* application decreased this increase. However, we did not observe any difference in terms of MDA levels in kidney tissue. We think that the results we obtained in the liver and heart tissue may be due to the effect of antioxidant membrane components, especially in *Chlorella*. The lack of difference in kidney tissue may be due to the longer time alcohol reaches these regions.

Thiol and GSH constitute the most important of the non-enzymatic intracellular antioxidants group among the advanced defense mechanisms to prevent cellular damage in the organism due to reactive oxygen species. The glutathione peroxidase system, one of the intracellular enzymatic antioxidants, includes the use of glutathione peroxidase enzymes, glutathione reductase (GR), reduced glutathione (GSH) and reduced NADPH as cofactors. Glutathione peroxidase; It reduces hydrogen peroxide and other organic peroxides by using GSH that is oxidized to glutathione disulfide forms (GSSG). They show activity either by inhibiting the formation of ROS or by scavenging free radicals and their precursors. In individuals with alcohol dependence, oxidant and antioxidant balances deteriorate and increase ROS[32,33].

Thanks to the antioxidants it contains, it is known that the protective effect of *Chlorella vulgaris* plays an active and preventive role in cases such as cell damage and cell death caused by oxidative stress-induced substances[34,35].

Chlorella vulgaris reduced the oxidative damage of lead, which is a heavy metal and has a very high toxic effect, in brain cells. In addition, hepatotoxicity caused by cadmium, which is a heavy metal and has a toxic effect, has been shown to have an inhibitory effect on tissue damage due to its antioxidant property[36].

In our study, we determined that ethyl alcohol caused a decrease in GSH-Px levels in the heart tissue, but this decrease was improved by the application of *Chlorella vulgaris*. We determined that *Chlorella vulgaris* did not change GSH-Px levels in liver and kidney tissues. We think that these changes may be due to the intracellular enzymatic system supportive activity of *Chlorella vulgaris* and the high antioxidant compounds it contains. In our literature review, we could not find any study showing the effect of *Chlorella vulgaris* on ethyl alcohol toxicity.

Alcohol use is associated with liver disease, neurotoxicity, hypertension, cardiomyopathy, immune response modulations, and an increased risk of cancer. In the studies, the effect processes of alcohol in such pathological conditions are not fully understood. The available data point out that the MAPK family plays a central role in these processes affected by alcohol[37,38,39].

In our study, it was determined that MAPK was increased in the liver in animals in the alcohol-administered group, but this alcohol-induced increase was suppressed in the *Chlorella*-treated group. However, in the literature review, no study was found on the effects of *Chlorella* on MAPK in alcohol toxicity. In our study, it can be thought that *Chlorella* has a corrective activity against liver damage caused by alcohol, with this aspect of liver MAPK levels increased by alcohol administration and decreased by *Chlorella* administration. In addition, the reasons for the alcohol-related decrease in MAPK activities in kidney and heart tissue require molecular evaluation.

As a result, it is evaluated that *Chlorella vulgaris* increases antioxidant enzyme levels especially in liver tissues against lipid peroxidation that occurs in tissues with the effect of alcohol, and has tissue protective activity with

this effect. In addition, increased MAPK activities in liver tissues as a result of alcohol intoxication improved with the effect of *Chlorella*, and it is thought that this situation may be associated with the protective effect of *Chlorella* on intracellular enzyme systems. New studies are still needed to determine the molecular pathway by which *Chlorella* displays its tissue protective activity in alcohol intoxication.

References

1. Muhtar N. Alkol Bağımlılarında Bağlanma. *İstanbul: İstanbul Üniversitesi Sağlık Bilimleri Enstitüsü, Psikiyatri Anabilim Dalı*. 2003.
2. Can M, Gürpınar SS, İşler H, Varol N, Gürpınarlı Z. Alkol Alan Kişilerin Kan Alkol Düzeyinin Solunum Havasındaki Alkol Düzeyi İle Karşılaştırılması Ve Karaciğer Enzimlerinin Etkisinin Araştırılması. Van Tıp Dergisi. 2008; 15(3): 75-80.
3. Andresen-Streichert H, Müller A, Glahn A, Skopp G, Sterneck M. Alcohol biomarkers in clinical and forensic contexts. Deutsches Ärzteblatt International; 2018. 115(18), 309.
4. Kültegin Ö, Defne T. Alkol ve Madde Kullanım Bozukluklarının Epidemiyolojisi. 3P Dergisi. 2003; 11(2): 123-128.
5. Chen L, Tang J, Song LN, Chen P, He J, Au CT et al. Heterogeneous photocatalysis for selective oxidation of alcohols and hydrocarbons. Applied Catalysis B: Environmental. 2019;242, 379-388.
6. Koob GF, Colrain IM. Alcohol use disorder and sleep disturbances: a feed-forward allostatic framework. Neuropsychopharmacology. 2020; 45(1), 141-165.
7. Palzes VA, Parthasarathy S, Chi FW, Kline-Simon AH, Lu Y, Weisner C, et al. Associations between psychiatric disorders and alcohol consumption levels in an adult primary care population. Alcoholism: clinical and experimental research. 2020;44(12), 2536-2544.
8. Scafato E, Caputo F, Patussi V, Balbinot P, Addolorato G, Testino G. The undertreatment of alcohol-related liver diseases among people with alcohol use disorder. Eur Rev Med Pharmacol Sci. 2020; 24(2), 974-82.
9. Crabb DW, Im GY, Szabo G, Mellinger JL, Lucey MR. Diagnosis and treatment of alcohol-associated liver diseases: 2019 practice guidance from the American Association for the Study of Liver Diseases. Hepatology. 2020; 71(1), 306-33.
10. Sonde V, D'souza A, Tarapore R, Pereira L, Khare MP, Sinkar P, et al. Simultaneous administration of diethylphthalate and ethyl alcohol and its toxicity in male Sprague-Dawley rats. Toxicology. 2000; (147) : 23-31.
11. Lackner C, Tiniakos D. Fibrosis and alcohol-related liver disease. Journal of hepatology. 2019;70(2), 294-304.
12. Sefarini M, Rio DD. Understanding the association between dietary antioxidants, redox status and disease: is the Total Antioxidant Capacity the right tool. Redox Report. 2004; 9(3). 145-152.
13. Bhatia S, Drake DM, Miller L, Wells PG. Oxidative stress and DNA damage in the mechanism of fetal alcohol spectrum disorders. Birth defects research. 2019; 111(12), 714-748.
14. Michalak A, Lach T, Cichoż-Lach H. Oxidative Stress—A Key Player in the Course of Alcohol-Related Liver Disease. Journal of clinical medicine. 2021; 10(14), 3011.
15. Żukowski, P, Maciejczyk M, Waszkiel D. Sources of free radicals and oxidative stress in the oral cavity. Archives of Oral Biology. 2018; 92, 8-17.
16. Engwa GA. Free radicals and the role of plant phytochemicals as antioxidants against oxidative stress-related diseases. Phytochemicals: Source of Antioxidants and Role in Disease Prevention. BoD–Books on Demand. 2018;7, 49-74.
17. Su LJ, Zhang JH, Gomez H, Murugan R, Hong X, Xu D, et al. Reactive oxygen species-induced lipid peroxidation in apoptosis, autophagy, and ferroptosis. Oxidative medicine and cellular longevity. 2019.
18. Morales M, Munné-Bosch S. Malondialdehyde: facts and artifacts. Plant physiology. 2019; 180(3), 1246-1250.
19. Lushchak VI, Lushchak O. Interplay between reactive oxygen and nitrogen species in living organisms. Chemico-Biological Interactions. 2021; 349, 109680.
20. Ursini F, Maiorino M. Lipid peroxidation and ferroptosis: the role of GSH and GPx4. Free Radical Biology and Medicine. 2020; 152, 175-185.
21. Alkadi H. A review on free radicals and antioxidants. Infectious Disorders-Drug Targets (Formerly Current Drug Targets-Infectious Disorders). 2020; 20(1), 16-26.
22. Yu M, Chen M, Gui J, Huang S, Liu Y, Shentu H, He J, Fang Z, Wang W, Zhang Y. (). Preparation of *Chlorella vulgaris* polysaccharides and their antioxidant activity in vitro and in vivo. International journal of biological macromolecules. 2019; 137, 139-150.
23. Abdel-Karim OH, Gheda SF, Ismail GA, Abo-Shady AM. Phytochemical Screening and antioxidant activity of *Chlorella vulgaris*. Delta Journal of Science. 2020; 41(1), 81-91.
24. Yue J, López JM. Understanding MAPK signaling pathways in apoptosis. International Journal of Molecular Sciences. 2020; 21(7), 2346.
25. Placer ZA, Cushman LL, Johnson BC. Estimation of product of lipid peroxidation (malonyl dialdehyde) in biochemical systems. Analytical biochemistry. 1966; 16(2), 359-364.
26. Sedlak J, Lindsay RH. Estimation of total, protein-bound, and nonprotein sulfhydryl groups in tissue with Ellman's reagent. Analytical biochemistry. 1968; 25, 192-205.
27. Lawrence RA, Burk RF. Glutathione peroxidase activity in selenium-deficient rat liver. Biochemical and biophysical research communications. 1976; 71(4), 952-958.
28. Jacob A, Wang P. Alcohol intoxication and cognition: implications on mechanisms and therapeutic strategies. Frontiers in neuroscience. 2020; 14, 102.
29. Gao S, Shi J, Wang K, Tan Y, Hong H, Luo Y. Protective effects of oyster protein hydrolysates on alcohol-induced liver disease (ALD) in mice: based on the mechanism of anti-oxidative metabolism. Food & Function. 2022;16.
30. Wu C, Liu J, Tang Y, Li, Ya, Q, Jiang Z. Hepatoprotective potential of partially hydrolyzed guar gum against acute alcohol-induced liver injury in vitro and vivo. Nutrients. 2019; 11(5), 963.
31. Fontes-Júnior EA, Maia CSF, Fernandes LMP, Gomes-Leal W, Costa-Malaquias A, Lima RR et al. Chronic alcohol intoxication and cortical ischemia: study of their comorbidity and the protective effects of minocycline. Oxidative Med. Cell. Longev. 2016; 1341453.

32. Carrard VC, Pires AS, Mendez M, Mattos F, Moreira JCF, Sant'Ana Filho M. Effects of acute alcohol consumption and vitamin E co-treatment on oxidative stress parameters in rats tongue. Food and chemical toxicology. 2009; 47(6), 1058-1063.

33. Wu D, Cederbaum AI. Oxidative stress and alcoholic liver disease. In Seminars in liver disease 2009; 29(02),141-154.

34. Mallick N. Copper-induced oxidative stress in the chlorophycean microalga Chlorella vulgaris: response of the antioxidant system. Journal of Plant Physiology. 2004;161(5), 591-597.

35. Shim JY, Shin HS, Han JG, Park HS, Lim BL, Chung KW, et al. Protective effects of Chlorella vulgaris on liver toxicity in cadmium-administered rats. Journal of Medicinal Food. 2008;11(3), 479-485.

36. Yun H, Kim I, Kwon SH, Kang JS, Om AS. Protective effect of Chlorella vulgaris against lead-induced oxidative stress in rat brains. Journal of Health Science. 2011; 57(3), 245-254.

37. Aroor AR, Shukla SD. MAP kinase signaling in diverse effects of ethanol. Life sciences. 2004; 74(19), 2339-2364.

38. Le Daré B, Lagente V, Gicquel T. Ethanol and its metabolites: update on toxicity, benefits, and focus on immunomodulatory effects. Drug metabolism reviews. 2019;51(4), 545-561.

39. Matyas C, Haskó G, Liaudet L, Trojnar E, Pacher P. Interplay of cardiovascular mediators, oxidative stress and inflammation in liver disease and its complications. Nature Reviews Cardiology. 2021; 18(2), 117-135.

Original Article
Eurasian Journal of Toxicology

Epidemiological and Clinical Investigation of Snake Bite Cases Admitted to the Emergency Department of a Tertiary Hospital in Izmir

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Abstract

Objectives: There are more than 3000 snake species in the world, about 30% of which are known to be venomous. Snake poisoning can cause serious mortality and morbidity. In this study, it was aimed to investigate epidemiologically and clinically the snake bite cases admitted to the emergency department of a tertiary hospital in Izmir.

Materials and Methods: Patients who applied to our emergency department with the complaint of snake bite between 2012 and 2022 were included in the retrospective observational study. The data of the patients were obtained by scanning the hospital electronic database. Vital signs, laboratory values, physical examination findings and outcomes of the patients were evaluated.

Results: Of the 52 patients, 42.3% were female and 57.7% were male, with a mean age of 48±16 years. It was seen that the most cases occurred in the summer season and only 1 patient died in total. Snake bite was seen on extremity in 41 (78.8%) patients while 11 (21.2%) had non-extremity bites. There was a statistically significant difference in creatine kinase (CK) value, K value, local edema and diffuse edema incidence between the two groups according to the bite site.

Conclusions: Higher CK, lower K level and more local and diffuse edema are seen in extremity bites compared to non-extremity bites.

Keywords: Snake bite, poisoning, environmental emergencies, emergency service.

Introduction

Poisoning due to snake bites is an important health problem all over the world. According to the World Health Organization (WHO), more than 5 million snake bites occur worldwide each year, with approximately 2.5 million poisonings and 81,000 to 138,000 deaths[1]. Approximately 30% of the 3000 snake species worldwide are venomous and considered dangerous to humans[2,3]. The most venomous species are grouped as Elapidae, Viperidae, Hydrophiida, Antractaspidida and Colubridae[4].

Of the snake species 41 are known in our country. 13 of these snakes are venomous. Of these venomous species, 10 are Viperidae (Vipers), 2 are Colubridae and one species is Elapidae. The Viperidae (viper) family is responsible for almost all of the snake bite cases in our country. The viper mostly causes hematotoxic effects, as well as local poisoning findings, necrosis in the skin and deep tissues[5]. Snake bites in Turkey are mostly seen in our Southern and Southeastern Anatolian region and especially in summer[6]. In addition, it is known that snake bites are mostly in the form of biting from the lower extremities[7]. It is known that correct and timely first aid and an effective treatment reduce the death rate, since snake venom can cause widespread effects on the bitten area and then on the whole body[8].

The first place of application for snake bites is emergency services and they cause serious poisoning that can result in mortality. For this reason, it is one of the diseases that must be managed well from the moment of admission to the emergency room. In this study, it was aimed to investigate the epidemiological and clinical features of snake bite cases admitted to the emergency department of a tertiary hospital in Izmir.

Methods

Study Design

This observational retrospective study was conducted between January 2012 and January 2022 in the emergency department of a tertiary university hospital. Vital signs and laboratory parameters of patients diagnosed with snake bite were analyzed. The patients were divided into 2 groups as with extremity and non-extremity bites. The 2 groups were compared in terms of vital signs, laboratory parameters, symptoms and outcome.

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Patients and Setting

Patients over the age of 18 who applied to the emergency department and were diagnosed with snake bite were included in the study. Patients whose results could not be reached, patients with missing data, pregnant women and those referred to an external center were excluded from the study.

Data Collection

The data of the patients included in the study were collected through the hospital information system and the vital parameters, demographic data and laboratory test results of the patients were recorded in the patient forms created to be used in the statistical analysis. The information about the exitus and discharge of the patients who were followed up for snake bites were noted. The obtained data were analyzed and compared between the 2 groups.

Statistical Analysis

SPPS 20 (IBM Corp. Released 2011. IBM SPSS Statistics for Windows, Version 20.0. Armonk, NY: IBM Corp.) statistical package program was used to evaluate the data. Mean ± standard deviation and Median (25% and 75% quartiles) percentage and frequency values were calculated for the variables. In addition, the homogeneity of the variances, which is one of the prerequisites of the parametric tests, was checked with the Levene test. Normality assumption was checked with the "Shapiro-Wilk" test. When it is desired to evaluate the differences between the two groups, "Student's t Test" if the parametric test prerequisites are met; if not, the "Mann Whitney-U test" was used. The relationship between two continuous variables was evaluated with the Pearson Correlation Coefficient, and if the parametric test prerequisites were not met, the Spearman Correlation Coefficient. The performance of a test can be defined by the test's diagnostic adequacy or its capacity to correctly classify cases into subgroups (healthy/patient etc.). Statistical significance level was accepted as $p < 0.05$ and $p < 0.001$.

Results

Between January 2012 and January 2022, 107 patients who applied to our emergency department and were diagnosed with snake bite were identified and 52 patients who met the criteria were included in the study. Exclusion criteria for the study are shown in the consult diagram (Figure 1).

A total of 52 patients were included in the study. Of these patients 22 (42.3%) were female and 30 (57.7%) were male and their mean age was 48±16. When the patients were divided into 2 groups according to the site of the snake bite, 41 (78.8%) patients were bitten from the extremity area, while 11 (21.2%) patients were bitten from the non-extremity area. Exitus developed in 1 (1.9%) patient and 51 (98.1%) patients were discharged. Descriptive statistics of the patients are presented in Table 1.

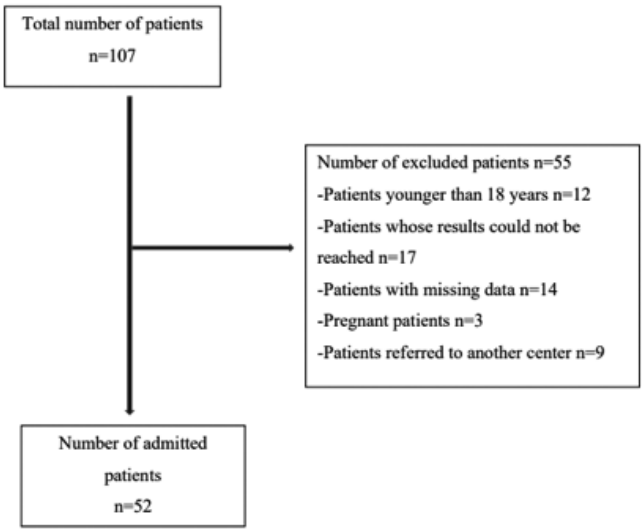

Figure 1: Consult diagram

Table 1: Descriptive statistics of patients

Location of the bite	Non-extremity	11(21.2)
	Extremity	41(78.8)
Tetanus vaccine	No	32(61.5)
	Yes	20(38.5)
Antivenom	No	27(51.9)
	Yes	25(48.1)
Analgesia	No	40(76.9)
	Yes	12(23.1)
Antibiotherapy	No	34(65.4)
	Yes	18(34.6)
Steroid	No	37(71.2)
	Yes	15(28.8)
Outcome	Ex	1(1.9)
	Discharge	51(98.1)

When the Creatine kinase (CK) and potassium (K) values of the patients were compared according to the bite sites, it was observed that the CK and K levels were statistically significantly higher in extremity bites ($p<0.05$). The comparison of the vital signs and laboratory results of the groups according to the snake bite site is presented in Table 2.

When the patients were compared in terms of the development of local edema, diffuse edema and systemic findings according to the bite sites, it was found that local edema and diffuse body edema were significantly more common in extremity bites ($p=0.001$, $p=0.002$ respectively). Considering the rate of development of systemic findings, it was seen that systemic findings developed only in 2 patients with non-extremity bites, and systemic findings did not develop in any of the extremity bites. These results show that there is no statistically significant difference between the groups ($p=0.212$) (Table 3).

Table 2: Comparison of vital signs and laboratory results of both groups and total patients

	Non-extremity (n = 11)	Extremity (n = 41)	Total (n = 52)	*P*
SBP	117 ± 15	120 ± 16	120 ± 16	.655
DBP	83 ± 8	76 ± 12	77 ± 12	.083
Pulse rate	78 ± 8	81 ± 9	81 ± 9	.269
WBC	12 ± 4	12 ± 5	12 ± 5	.831
Neutrophil	9 ± 5	10 ± 5	10 ± 5	.663
Lymphocyte	1.3 ± 0.9	1.50 ± 1.08	1.50 ± 1.05	.519
Platelet	215000 ± 91000	196000 ± 82000	200000 ± 83000	.496
INR	1.04±0.11	1.10 ± 0.19	1.09 ± 0.18	.300
APTT	26 ± 4	30 ± 8	29 ± 8	.130
Creatine (mg/dl)	0.84 ± 0.13	0.81 ± 0.16	0.82 ± 0.15	.577
CK (U/L)	61 ± 28	374 ± 458	308 ± 425	.029
K (mmol/L)	4.50 ± 0.48	4.13 ± 0.52	4.21 ± 0.53	.038

SBP: Systolic blood pressure, SBP:diastolic blood pressure, WBC:White blood cell, INR :international normalized ratio, APTT :activated partial thromboplastin time, CK: creatine kinase, K: potassium

Table 3: Development of local edema, diffuse edema and systemic findings according to the bite site

		Bite location		*P*
		Non-extremity	Extremity	
Local edema	**No**	10 (90.9%)	12 (29.3%)	***.001***
	Yes	1 (9.1%)	29 (70.7%)	
	Total	11(100%)	41(100%)	
Comman edema	**No**	10 (90.9%)	16(39%)	***.002***
	Yes	1 (9.1%)	25(61%)	
	Total	11(100%)	41(100%)	
Systemic findings	**No**	9 (81.8%)	41(100%)	***.212***
	Yes	2 (18.2%)	0(0%)	
	Total	11(100%)	41(100%)	

Considering the months in which snake bites occur during the year, it is seen that there is an increase in the number of cases in the 7th, 8th and 9th months of the year. The distribution of the number of cases by months is presented in Figure 2.

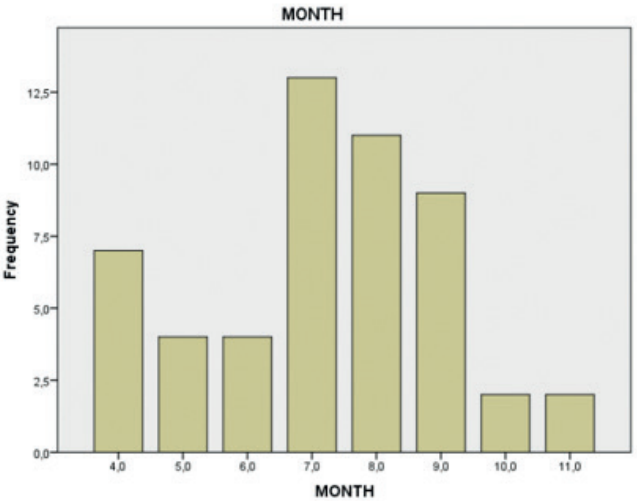

Figure 2: Distribution of the number of cases by months.

Discussion

In this study, snake bites seen in the Izmir region were evaluated, and the majority of the cases were seen in the summer season. Extremity bites, on the other hand, were associated with the development of local and diffuse edema, and elevated CK and K levels.

It is known that snake venom can prolong thrombocytopenia, leukocytosis, prothrombin time and partial thromboplastin time with its procoagulant effect, and it has been reported that these laboratory changes are associated with poor prognosis and mortality[9]. In a study by Gutiérrez et al., it was reported that there was an increase in White blood cell (WBC) count and neutrophil values, which indicate the inflammatory response, due to snake bite[10]. Oto et al. In their study, in which they aimed to present the clinical course, complications and treatment approaches of the patients, the platelet value was found within the reference values determined for normal population, and the results are compatible with our study[11]. Moon et al. In a study conducted by international normalized ratio (INR) and activated partial thromboplastin time (APTT), it was stated that there may be a prolongation in snake bite[12]. In this study, it was observed that there was an increase in WBC and neutrophil values due to snake bite, in accordance with the literature, and there was no significant change in PLT, INR and APTT values, contrary to the literature. The absence of coagulopathy findings in our study may be related to regional snake venom characteristics. However, there are not enough studies on this subject in the Izmir region. It is known that there may be an increase in serum CK, K, and myoglobin in snake bites due to rhabdomyolysis[13]. In this study, snake bite was found not to be associated with an increase in K, but was associated with an increase in CK. In addition, when we classify the patients according to the bite sites, it is seen that CK and K levels are significantly higher in extremity bites than in trunk bites. Considering that the reason for the increase in CK and K is rhabdomyolysis, the denser muscle tissue of the extremities and the thinner subcutaneous fat tissue in the extremities compared to the trunk may explain this difference. The clinical effects of snake bites can range from mild local reactions to life-threatening systemic reactions. Local tissue edema may occur as a result of the direct effect of the toxin on the tissues and the pressure during the bite[14]. In our study, when local edema and diffuse edema development were compared between bites with and without extremities, it was seen that both local edema and diffuse body edema were more common in bites in the extremities. In addition, it was observed that systemic

findings developed in only two patients with trunk bite and no systemic findings developed in any extremity bite. Early treatment and antivenom applications are closely related to survival[15]. In addition to antivenom therapy, tetanus prophylaxis, antibiotic therapy, extremity elevation and rest splint are recommended for snake bites[16]. In recent studies, antibiotics are recommended instead of routine antibiotics in cases such as tissue necrosis, bullae, abscess development in the bitten area, and inappropriate first aid intervention (such as incision, suction)[17,18]. Antivenom was administered to 48.1% of our patients, and all of these patients survived. In the study, it is seen that antivenom treatment was not applied to the only patient who died. Antibiotherapy was administered to 34.6% of the patients in our study, which is consistent with the literature recommendations. It is known that snake bites occur mostly in the summer period[6]. In this study, snake bites occurred at a much higher rate in July, August and September compared to other months. This situation can be explained by the increase in the number of workers working in the rural areas during the summer months and the increase in the possibility of encountering snakes.

This study has certain limitations. The primary limitation of the study is that it was single-centered and carried out with a small number of patients. In addition, due to the retrospective nature of the study, some inaccessible data could not be evaluated. Prospective studies with a larger patient population are needed.

Conclusion

Our study is, to our knowledge, the first to compare snake bites with and without limbs. Local edema and diffuse edema are more common in bites from the extremity. Bites from the extremities have a higher CK value and a lower K value. There is a need for multicenter studies with more patients on this subject.

References

1. WHO. Snakebite envenoming- a strategy for prevention and control. May 2, 2019. Available at https://apps.who.int/iris/rest/bitstreams/1216706/retrieve.
2. Sahyoun C, Rima M, Mattei C, Sabatier JM, Fajloun Z, Legros C. Separation and analytical techniques used in snake venomics: A reviewe article. Processes 2022;10(7):1380.
3. Baran İ, Başoğlu M. Türkiye Sürüngenleri, Kısım 2, Yılanlar Ege Üniversitesi. Basımevi, İzmir, 1998:9-25.
4. Kose R. [The management of snake envenomation: evaluation of twenty-one snake bite cases]. Ulus Travma Acil Cerrahi Derg. 2007;13(4):307-12.
5. Okur Mİ, Yıldırım AM, Köse R. Venomous snake bites and its therapy in Turkey. Turkiye Klinikleri Journal of Medical Sciences 2001; 21: 528.
6. Gökel Y, Başlamışlı F, Koçak R. Çukurova yöresinde yılan ısırmaları. Çukurova Üniversitesi Tıp Fakültesi Dergisi 1997;22:184-8.
7. Garg A, Sujatha S, Garg J, Acharya NS, Chandra Parija S. Wound infections secondary to snakebite. J Infect Dev Ctries. 2009; 3:221–23.
8. Warrell DA. Treatment of snakebite in the Assia Passific: A personal view. In: Gopalaksishnakone P, Chou LM, eds. Snake of medical. basım yeri bilinmiyor: importance singapore venom ad. Toxin Research Group. National University of Singapore. 1990; 641-70.
9. Benvenuti LA, Franca FO, Barbaro KC,Nunes JR, Cardoso JL. Pulmonary haemorrhage causing rapid death after Bothrops jararacussu snakebite: a case report. Toxicon 2003;42:331-4.
10. Gutiérrez, JM, Calvete JJ, Habib AG, Harrison RA, Williams DJ, Warrell DA. (2017). Snakebite envenoming. Nature reviews. Disease primers, 3, 17063. https://doi.org/10.1038/nrdp.2017.63.
11. Oto A, Hapolat YK, Venomous Snakebites in Children in Southeast Turkey. Dicle Med J 2021; 48(4): 761-9.
12. Moon JM, Chun BJ, Cho YS, Kim JC, Koo YJ, Park KH et al. Coagulopathy after snake envenomation in South Korea. Clin Toxicol (Phila). 2021;59(10):905-12. doi: 10.1080/15563650.2021.1884694.
13. Warrell DA. Venomous Bites, Stings, and Poisoning: An Update. Infect Dis Clin North Am. 2019 Mar;33(1):17-38. doi: 10.1016/j.idc.2018.10.001.
14. Satar S, Karcıoğlu Ö, Sebe A. An usual localization of snakebite treated without antivenin: case report. The Mount Sınai J of Med 2005; 72:116-9.
15. Yüksel M, Eryiğit V, Erimşah ME, Karaaslan U, Büyükyılmaz T, Ersan E. The Attitudes of Emergency Physicians in Turkey towards the Snakebites. Kocaeli Med J 2020; 9; 1:76-82.
16. Seğmen MS, Acehan S, Avcı A, Gülen M, Satar S. Demographic and Clinical Characteristics of Patients Presenting to the Emergency Department with Snakebite. Phnx Med J 2020; 2(2):72-8.
17. Tagwireyi DD, Ball DE, Nhachi CF. Routine prophylactic antibiotic use in the management of snakebite. BMC Clin Pharmacol. 2001;1:4. doi: 10.1186/1472-6904-1-4.
18. Clark RF, Selden BS, Furbee B. The incidence of wound infection following crotalid envenomation. J Emerg Med. 1993;11:583-6.

Original Article

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Frontal QRS-T Angle in Scorpion Stings

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Abstract

Introduction: The aim of this study was to investigate the change in frontal QRS-T angle in different clinical stages of scorpion stings.

Materials-Methods: In this retrospective study, laboratory data of patients and T angle, QRS duration (ms), QT duration (ms), and QTc duration (ms) of the patients who presented with scorpion sting were calculated and recorded in the data file. The results were analyzed.

Results: Eighty patients who applied to the emergency department with the complaint of scorpion sting were included in our study. Forty-four patients were evaluated as Stage I, 26 as Stage II, and 10 as Stage III. The patient groups did not differ in age ($p = 0.605$) and sex ($p = 0.432$). No significant difference was observed between the laboratory findings of the patients at the time of admission. ECG findings showed a considerable difference in frontal QRS-T angles between scorpion sting stages ($p < 0.001$). Pairwise comparison of the stages with post-hoc analysis revealed a non-significant difference between Stages I and II ($p = 0.143$), and a significant difference between Stages I and III ($p < 0.001$) and Stages II and III ($p = 0.003$). Correlation analysis results showed that the frontal QRS-T angle was negatively correlated with age ($r = -0.281$, $p = 0.016$) and positively correlated with the clinical stage ($r = 0.384$, $p = 0.001$). Multivariate linear regression analysis was performed to identify independent predictors of frontal QRS-T angle, and the stage of the scorpion sting was identified as an independent predictor ($p = 0.001$).

Conclusions: The increase in frontal QRS-T angle in scorpion stings may be used as a parameter that can help both early detections of cardiac involvement and clinical staging.

Keywords: Emergency department, Arthropods, Scorpion sting, frontal QRS-T angle, Electrocardiography.

Introduction

Scorpion stings are more common in subtropical and tropical countries, as climatic conditions, drought, and temperature are important risk factors[1]. Scorpions are venomous arthropods that are members of the class Arachnida and the order Scorpiones. Of the 2700 described species, 30-50 species are poisonous to humans, and most of these species are in the genera *Buthus*, *Parabuthus*, *Mesobuthus*, *Tityus*, *Leiurus*, *Androctonus*, or *Centruroides*, which belong to the Buthidae family. Of these genera, *Androctonus*, *Leiurus*, and *Mesobuthus* are the most medically important scorpion species in Turkey[2]. A scorpion sting can lead to variable and complex clinical manifestations, ranging from a local effect to intense autonomic nervous system responses and a systemic inflammatory reaction. These symptoms often progress to serious cardiac and pulmonary changes that can be fatal, especially in children and the elderly[3,4].

Some electrocardiography (ECG) findings are also observed following scorpion stings. Sinus tachycardia, sinus bradycardia, biphasic and notched T wave, as well as electrical alternans, prolonged QT interval, ST elevation or depression, albeit rare, are ECG abnormalities that can be observed following in scorpion stings[5]. Another parameter obtained in ECG evaluation is the frontal-QRS-T angle. The frontal QRS-T angle is an alternative to the spatial QRS-T angle and can be easily calculated from the anterior QRS axis and T wave axis on a 12-lead electrocardiogram[6]. Reportedly, abnormal spatial and frontal QRS-T angle was associated with both mortality and coronary heart disease[7]. Furthermore, studies have reported that frontal QRS-T angle is associated with non-arrhythmic or sudden cardiac death and risk of cardiovascular disease in the general population[8]. Although, to the best of our knowledge, no study in the literature has investigated QRS-T angle in scorpion stings, QRS-T angle reportedly increases in cases of hypertension, conversive attack, chronic obstructive pulmonary disease, and nonfunctional adrenal adenomas[9-12].

The aim of the present study was to investigate the changes in frontal QRS-T angle and other ECG parameters according to clinical stages of patients who presented to the emergency department with the complaint of scorpion sting.

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Materials and Methods

Patient selection

This retrospective preliminary study was initiated after approval was obtained from the university ethics committee (HRÜ/22.17.23).

Patient records were scanned in the hospital electronic record system with the ICD-10 codes X22 (contact with scorpions) and T63.2 (toxicity of scorpion venom). A patient data file containing basic characteristics such as age and sex, laboratory results, and ECG parameters was created. Patients whose files and ECG records could not be accessed, patients with bundlebranch block and ventricular hypertrophy on ECG, patients with atrial fibrillation, coronary artery disease, patients with previous coronary bypass and heart valve surgery history, and those under 18 years of age were excluded from the study. Patients over 18 years of age who did not meet the exclusion criteria were included in the study. The patients included in the study were clinically divided into four stages[13].

Stage I: Mild pain or paresthesia in the bitten area, no systemic findings
Stage II: Very severe pain or paresthesia extending outside the bitten area (one entire extremity), severe pain on touch.
Stage III: Somatic neuromuscular or cranial nerve involvement findings such as jerks in the extremities, involuntary tremor, cranial nerve involvement (blurred vision, eye movement disorder, salivation, tongue fasciculations, dysphagia, and speech disorder, among others),
Stage IV: Presence of both somatic neuromuscular and cranial nerve involvement. In addition, organ failure findings such as myocardial infarction, pulmonary edema, convulsions, shock

ECG measurements

A 12-lead surface ECG was recorded for all patients with a paper speed of 25 mm/s and an amplitude of 10 mm/mV. To minimize calculation errors, all ECGs were transferred to a digital platform and measurements were made under a magnifying glass. PR duration was calculated from the beginning of the P wave to the beginning of the QRS complex. QRS duration was calculated from the onset of QRS. The corrected QT interval (QTc) was calculated according to Bazett's formula: QTc = QT/√RR. QRS axis and T axis data were obtained through automated reports of ECG recordings and frontal QRS-T angle was obtained using the formula QRS axis - T axis.

Laboratory Measurements

White blood cell (3.7–10.1 10e3/μL), hemoglobin (12–18 g/dL), hematocrit (35%–53.7%), and platelet (142–424 10e3/μl) counts were determined with the Alinity HQ (Abbott, USA). Serum glucose (70–105 mg/dL), urea (10–50 mg/dL) and creatinine (0.2–1.11 mg/dL) levels were measured by conventional laboratory methods on Atellica Solution (Siemens Healthineers, Germany).

Statistical analysis

The data was statistically analyzed using the Statistical Package for the Social Sciences v.21.0 (IBM Corporation, Armonk, NY, USA) software package. Shapiro–Wilk test was used to test the normality hypothesis for the distribution of the continuous variables. Non-normally distributed data were expressed as Median (IQR: interquartile range), whereas qualitative data were expressed as percentage values. Kruskal–Wallis test was used to compare the five groups. Pairwise comparisons were then performed using Dunn's post-hoc test and the Chi-Square (cross-tab) was used to compare the categorical data. Pearson correlation analysis was performed to investigate the correlation between the data. After correlation analysis, linear regression analysis was performed with the stepwise method. $P < 0.05$ was considered statistically significant in all analyses.

Results

Eighty patients who applied to the emergency department with the complaint of scorpion sting were included in our study. Forty-four of the patients were evaluated as Stage I, 26 as Stage II, and 10 as Stage III. There was no difference in terms of age ($p = 0.605$) and sex ($p = 0.432$) between the patient groups. There was no significant difference between the laboratory findings of the patients at the time of admission (Table 1). Most commonly stung areas were upper extremity (47.9%, n = 35), lower extremity (38.4%, n = 28), torso (11%, n = 8), and head and neck region (2.7%, n = 2). All of the patients were discharged in good health and no patient died.

ECG findings obtained at the time of admission showed no significant difference in QRS and QT durations, and despite the presence of a difference between QTc values ($p = 0.021$), it was found to be between 360–440 ms. A significant difference was observed in frontal QRS-T angles between different scorpion sting stages ($p < 0.001$). Pairwise comparison of the stages with post-hoc analysis revealed a non-significant difference between Stage I and Stage II ($p = 0.143$), and a significant difference between Stage I and Stage III ($p < 0.001$) and Stage II and Stage III ($p = 0.003$) (Figure 1).

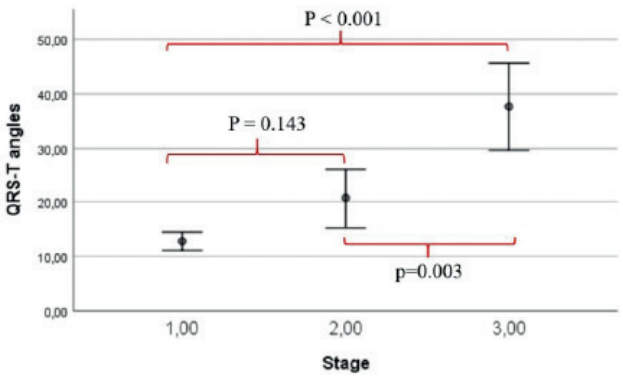

Figure 1:

Table 1: Basic data according to clinical stages of scorpion sting

	STAGE I	STAGE II	STAGE III	P
N (F/M)	44 (24/20)	26 (12/14)	10 (7/3)	0.432
Age (year)	35.50 (27.00–46.00)	33.00 (26.00–40.75)	40.50 (18.00–49.50)	0.605
Systolic blood pressure (mmHg)	120.00 (115.00–144.00)	135.00 (113.75–142.50)	125.00 (90.00–132.50)	0.371
Diastolic blood pressure (mmHg)	78.00 (71.00–87.00)	80.00 (75.25–90.00)	70.00 (56.00–82.50)	0.039
Pulse rate (pulse/minute)	84.00 (73.00–96.00)	80.00 (73.25–88.50)	71.50 (66.00–91.75)	0.385
White blood cell (10e3/µL)	7.56 (6.63–8.20)	7.80 (7.09–8.56)	9.00 (5.88–9.62)	0.422
Hemoglobin (g/dL)	14.00 (12.70–14.60)	13.10 (12.13–14.00)	12.30 (11.90–14.80)	0.032
Glucose (mg/dL)	97.00 (86.00–105.00)	95.00 (84.75–122.50)	108.00 (101.00–115.75)	0.166
Urea (mg/dL)	27.80 (19.20–32.10)	23.00 (14.39–30.43)	25.60 (18.50–30.50)	0.370
Creatinine (mg/dL)	0.70 (0.60–0.80)	0.60 (0.58–0.70)	0.60 (0.57–0.73)	0.304
Potassium	4.00 (3.80–4.30)	4.00 (3.98–4.50)	3.84 (3.70–4.21)	0.361
C-Reactive Protein	0.20 (0.08–0.47)	0.16 (0.05–0.35)	0.25 (0.00–0.91)	0.510
Frontal QRS-T angle	12.00 (8.00–18.00)	16.00 (10.25–37.50) [a]	41.50 (22.00–47.00)[bc]	<0.001
QRS duration (ms)	86.00 (80.00–96.00)	84.00 (78.00–92.50)	90.00 (85.00–95.00)	0.449
QT duration (ms)	398.00 (360.00–434.00)	364.00 (362.00–388.00)	388.00 (368.50–416.00)	0.142
QTc duration (ms)	428.50 (403.00–450.00)	412.00 (399.25–418.25)	415.00 (398.25–422.00)	0.021

a: p = 0.143 when comparing Stage I with Stage II, b: p < 0.001 when comparing Stage I with Stage III, c: p = 0.003 when comparing Stage II with Stage III

Table 2: Correlation analysis results between frontal QRS-T angle and other parameters

	Frontal QRS-T angle	
	r	p
Age (year)	−0.281	0.016
Systolic blood pressure (mmHg)	−0.164	0.165
Diastolic blood pressure (mmHg)	−0.149	0.209
Pulse rate (pulse/minute)	−0.045	0.706
White blood cell (10e3/µL)	0.228	0.053
Hemoglobin (g/dL)	−0.086	0.467
Glucose (mg/dL)	−0.028	0.817
Urea (mg/dL)	0.137	0.248
Creatinine (mg/dL)	0.082	0.491
Potassium	0.199	0.092
C-Reactive Protein	−0.169	0.152
QRS duration	−0.188	0.111
QT interval	0.017	0.886
Corrected QT interval (QTc)	−0.03	0.798
Clinical stage	0.384	0.001

Correlation analysis results showed that frontal QRS-T angle was negatively correlated with age (r = −0.281, p = 0.016) and positively correlated with clinical stage (r = 0.384, p = 0.001). The correlation results between the frontal QRS-T angle and the data are given in Table 2. Multivariate linear regression analysis was performed to identify independent predictors of frontal QRS-T angle, and the stage of scorpion sting was identified as an independent predictor (p = 0.001) (Table 3).

Table 3: Multivariate linear regression analysis showing independent predictors of frontal QRS-T angle

	Unstandardized Coefficients		Standardized Coefficients			
	B	Std. Error	Beta	t	P	95 % Confidence Interval
(Constant)	6.440	2.926		2.201	0.031	0.606–12.274
Clinical stage	6.627	1.890	0.384	3.507	0.001	2.859–10.396
R2 = 0.148, R2 (Adjusted) = 0.136 Model p = 0.001						

Discussion

In the present study, it was found that the frontal QRS-T angle was significantly increased in Stage III scorpion sting patients compared to Stage I and Stage II, but no significant difference was found between Stage I and Stage II patients. In addition, clinical stage of scorpion sting appeared to be an independent predictor of frontal QRS-T angle.

Venom glands and stings of scorpions are located in their curved tails. Most scorpion stings lead to the development of local symptoms, including intense pain in the skin, followed by hyperemia, scarring, and itching after venom inoculation[2]. Hyaluronidases and other enzymes subsequently increased tissue permeability and the toxins reaching circulation spread throughout the body. Circulating toxins accumulate in kidney, blood, liver, lung, heart, and spleen, causing various effects[4,14]. These effects lead to a mixed cholinergic/adrenergic toxidrome with massive release of epinephrine,

norepinephrine and other vasoactive peptides[15]. Lung and heart were affected immediately after injection owing to high blood supply[4]. With the effect of the toxin on the heart and lungs, serious cardiac complications such as myocardial infarction, acute pulmonary edema, cardiogenic shock, myocarditis, and even death can occur[16,17]. QRS-T angle is defined as the angle between the ventricular depolarization and repolarization directions. Therefore, a wide QRS-T angle reflects structural abnormalities that affect depolarization or regional pathophysiological changes in ionic channels that change the repolarization order. In cases when there is an imbalance in the electrical activation and recovery of the ventricles, the QRS axis and T wave axis are no longer aligned and the QRS-T angle widens. Widening of the frontal QRS-T angle was associated with mortality mainly in patients with narrow QRS without bundlebranch block[7] or is a predictor of ventricular arrhythmia[18,19]. Furthermore, the frontal QRS-T angle is reportedly an indicator of arrhythmic events in patients with reduced left ventricular function[18]. However, Aro LA et al. evaluated the ECG recordings of middle-aged patients and found that wide QRS-T angle was associated with arrhythmic mortality; however, QRS-T angle did not predict non-arrhythmic deaths[8].

In scorpion stings, cardiac involvement and ECG changes occur through many mechanisms[5]. Scorpion venom is a water-soluble antigenic complex and acts as a mixture of neurotoxin, cardiotoxin, nephrotoxin, hemolysins, phosphodiesterases, and histamine[20]. Cardiac outcomes such as myocardial damage, pulmonary edema, and cardiogenic shock may occur as a result of the release of vasoactive, inflammatory and thrombogenic peptides and amine components (histamine, serotonin, bradykinin, leukotrienes, and thromboxane) that act on the coronary vasculature and induce coronary artery vasospasm and facilitate platelet aggregation as well as thrombosis16. In addition, scorpion venom inhibits the angiotensin converting enzyme, which leads to the accumulation of bradykinin, which plays a role in the development of pulmonary edema[20]. Another mechanism for the cardiac effects of scorpion stings may be immunoglobulin E-mediated immediate hypersensitivity to scorpion venom in susceptible individuals[21]. In summary, early cardiac dysfunction is associated with the "vascular phase" of scorpion poisoning. This leads to increased left ventricular afterload, impaired left ventricular discharge, increased left ventricular filling pressure and capillary critical pressure, which is characterized by deep catecholamine-related vasoconstriction, resulting in pulmonary edema and increased right ventricular afterload[22]. To the best of our knowledge, there are no studies in the literature that have evaluated frontal QRS-T angles in scorpion stings. As the clinical staging of the scorpion sting increases, the systemic effects of the venom begin to appear. An initial increase in blood pressure and cardiac output, followed by decreased left ventricular function and hypotension may occur. The reason for the increase in frontal QRT-T angle in patients suffering from advanced stages of scorpion stings is not fully known. Although there were no Stage IV patients who developed organ failure in the present study, one of the reasons for the increase in frontal QRS-T angle in Stage III patients may be the intense release of epinephrine, norepinephrine, and other vasoactive peptides that occur after scorpion stings. In addition, changes in blood pressure after scorpion stings may also contribute to the increase in frontal QRT-T angle. In fact, it has been reported that abnormal QRS-T angle is a risk factor for hypertension in Type 2 DM patients[9]. In addition, nondipping hypertension has been associated with an increased frontal QRS-T angle, and it has been suggested that increased blood pressure during sleep may affect cardiac repolarization[23]. QRS-T angle is also used to monitor response to treatment in hypertension. Elffers et al. evaluated the effects of antihypertensive treatment using ECG and found that the QRS-T angle decreased with treatment but was not anatomically associated with a reduction in left ventricular hypertrophy[24]. Although no Stage IV patients were present in the current study, we think that the increase in frontal QRS-T angle due to blood pressure changes seen in the early stages may provide preliminary information about possible cardiac involvement.

Limitations

The main limitation of the present study was that it was conducted in a single-center and the number of patients was relatively small. Although Androctonus and Leiurus are known to be the most medically important scorpion species in our region, the individual scorpion species that the patients in our study were exposed to could not be determined. In addition, the ECG recordings of the patients before the scorpion sting could not be obtained and the baseline frontal QRS-T angles were not known. Another limitation of the study is that it was designed retrospectively, the patients did not have post-treatment ECG recordings, and the change in frontal QRS-T angles with treatment could not be evaluated. These limitations in our preliminary study will guide and provide insights for future studies.

Conclusion

In conclusion, the increase in frontal QRS-T angle in scorpion stings may be used as a parameter that can help both early detection of cardiac involvement and clinical staging.

References

1. Santos MS, Silva CG, Neto BS, Grangeiro Júnior CR, Lopes VH, Teixeira Júnior AG, et al. Clinical and Epidemiological

Aspects of Scorpionism in the World: A Systematic Review. Wilderness Environ Med 2016;27:504-518.
2. Celik E, Caglar A, Celik SF. Clinical Effects and Predictive Factors Affecting the Clinical Severity of Scorpion Envenomations in Western Turkey. J Trop Pediatr 2021;67.
3. Fukuhara YD, Reis ML, Dellalibera-Joviliano R, Cunha FQ, Donadi EA. Increased plasma levels of IL-1beta, IL-6, IL-8, IL-10 and TNF-alpha in patients moderately or severely envenomed by Tityus serrulatus scorpion sting. Toxicon 2003;41:49-55.
4. Isbister GK, Bawaskar HS. Scorpion envenomation. N Engl J Med 2014;371:457-463.
5. Gueron M, Ilia R, Sofer S. The cardiovascular system after scorpion envenomation. A review. J Toxicol Clin Toxicol 1992;30:245-258.
6. Oehler A, Feldman T, Henrikson CA, Tereshchenko LG. QRS-T angle: a review. Ann Noninvasive Electrocardiol 2014;19:534-542.
7. Zhang ZM, Prineas RJ, Case D, Soliman EZ, Rautaharju PM, ARIC Research Group. Comparison of the prognostic significance of the electrocardiographic QRS/T angles in predicting incident coronary heart disease and total mortality (from the atherosclerosis risk in communities study). Am J Cardiol 2007;100:844-849.
8. Aro AL, Huikuri HV, Tikkanen JT, Junttila MJ, Rissanen HA, Reunanen A, et al. QRS-T angle as a predictor of sudden cardiac death in a middle-aged general population. Europace 2012;14:872-876.
9. Cardoso CR, Leite NC, Salles GF. Factors associated with abnormal T-wave axis and increased QRS-T angle in type 2 diabetes. Acta Diabetol 2013;50:919-925.
10. Ocak M, Tascanov MB, Kemer Z, Yasak İH, Fedai Ü. Examination of ECG characteristics of patients admitted to emergency department with conversive attack. Am J Emerg Med 2021;48:110-113.
11. Hocanli I, Tanriverdi Z, Kabak M, Gungoren F, Tascanov MB. The relationship between frontal QRS-T angle and the severity of newly diagnosed chronic obstructive pulmonary disease. Int J Clin Pract 2021;75:e14500.
12. Topaloglu O, Cimci M. The frontal QRS-T angle in patients with incidentally discovered nonfunctional adrenal adenomas. Eur Rev Med Pharmacol Sci 2021;25:3028-3037.
13. HI H. Arthropods. 10 ed. New York: Mc Graw Hill Education, 2015.
14. Reis MB, Zoccal KF, Gardinassi LG, Faccioli LH. Scorpion envenomation and inflammation: Beyond neurotoxic effects. Toxicon 2019;167:174-179.
15. Ismail M. The treatment of the scorpion envenoming syndrome: the Saudi experience with serotherapy. Toxicon 1994;32:1019-1026.
16. Bahloul M, Chaari A, Dammak H, Samet M, Chtara K, Chelly H, et al. Pulmonary edema following scorpion envenomation: mechanisms, clinical manifestations, diagnosis and treatment. Int J Cardiol 2013;162:86-91.
17. Bayar N, Küçükseymen S, Yüksel IO, Arslan S. [Rapidly improving acute myocarditis after a scorpion sting]. Turk Kardiyol Dern Ars 2013;41:629-632.
18. Borleffs CJ, Scherptong RW, Man SC, van Welsenes GH, Bax JJ, van Erven L, et al. Predicting ventricular arrhythmias in patients with ischemic heart disease: clinical application of the ECG-derived QRS-T angle. Circ Arrhythm Electrophysiol 2009;2:548-554.
19. Kurisu S, Nitta K, Watanabe N, Ikenaga H, Ishibashi K, Fukuda Y, et al. Effects of Newly Developed Right Versus Left Bundle Branch Block on the QRS Axis, T-wave Axis and Frontal QRS-T Angle in Patients with a Narrow QRS. Intern Med 2021;60:25-30.
20. Bawaskar HS, Bawaskar PH. Scorpion sting. J Assoc Physicians India 1998;46:388-392.
21. Kounis NG. Kounis syndrome (allergic angina and allergic myocardial infarction): a natural paradigm? Int J Cardiol 2006;110:7-14.
22. Abroug F, Souheil E, Ouanes I, Dachraoui F, Fekih-Hassen M, Ouanes Besbes L. Scorpion-related cardiomyopathy: Clinical characteristics, pathophysiology, and treatment. Clin Toxicol (Phila) 2015;53:511-518.
23. Tanriverdi Z, Unal B, Eyuboglu M, Bingol Tanriverdi T, Nurdag A, Demirbag R. The importance of frontal QRS-T angle for predicting non-dipper status in hypertensive patients without left ventricular hypertrophy. Clin Exp Hypertens 2018;40:318-323.
24. Elffers TW, Trompet S, de Mutsert R, Maan AC, Lamb HJ, Macfarlane PW, et al. Electrocardiographic Detection of Left Ventricular Hypertrophy; Adding Body Mass Index and Spatial QRS-T Angle: A Cross-Sectional Study. Cardiol Ther 2019;8:345-356.

Review Article

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Electrocardiography and Drug Intoxication

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Abstract

Intoxication is the deterioration of body functions due to different toxic substances. Poisoning by drugs constitutes an important part of all poisonings. Symptoms such as altered consciousness, tachycardia/bradycardia, or hypertension/hypotension may be seen because the cardiovascular system is affected. Changes in clinical findings and ECG may be revealed according to the degree of heart involvement. Rapid recognition and effective intervention by the emergency physician are of great importance. This review considers the use of ECG in the management of poisoned patients. Systematic evaluation of the ECG in a patient followed up with poisoning is essential for details that may be overlooked. Velocity, rhythm, intervals, and segments, QRS, wave morphologies, durations, ischemic changes should be followed carefully.

When performing rhythm analysis, clues to drug cardiotoxicity should be sought in unstable patients. Are there ectopic beats on the ECG? The answer to this question may carry important clues. Automaticity caused by sympathomimetics may underlie ectopic beats. This may be the first sign of a problem caused by acute coronary syndrome or electrolyte disturbances. Is the rhythm supraventricular? or ventricular? Is bradycardia with AV block? Or without AV block? Is tachycardia narrow complex? Or is it a large complex? Answers to questions such as: For life-threatening rhythms, ventricular tachycardia, ventricular fibrillation, and complete AV-block, the guidelines developed should be followed, and first intervention should be made. Agents that can cause tachycardia; are sympathomimetics (methamphetamine), anticholinergics (antidepressants, antipsychotics), class 1A and 1C antidysrhythmics, and TCA. Agents that can cause bradycardia; calcium channel / beta blockers / digoxin (AV block), opioids / ethanol, organophosphates, lithium. Prolonging the PR interval may indicate beta-adrenergic antagonism, calcium channel antagonism, or digoxin poisoning. Typical ECG of TCA poisoning shows sinus tachycardia with first-degree AV block, wide QRS complexes, and positive R' wave in aVR. The ECG should be taken and evaluated in patients presenting with poisoning within the first 10 minutes. Suppose the poisoning agent is an agent that influences the cardiovascular system. In that case, it should be kept in mind that continuous cardiac monitoring and control ECG evaluation should be performed in addition to the application of ECG.

Keywords: Intoxication, Electrocardiography, Cardiotoxic

Introduction

Substances that cause death with their chemical and physiological effects when taken in toxic doses are called toxins[1]. Intoxication is the deterioration of body functions due to different toxic substances[2]. This can affect the person's vital functions. Depending on the causes of poisoning, various symptoms can be seen in the person. Poisonings: In terms of the way it occurs, it can be environmental, industrial, home accidents, occupational, suicidal, murder accidental, and because of an attack. The annual incidence of poisoning in our country is between 0.8-5%3. When the applications made are examined, poisoning cases are mostly seen in the 0-6 age group among children and 17-29 years old among adults4. The mortality and morbidity of poisonings are high. This rate ranges from 0.3% to 27%[5]. Poisoning by drugs constitutes an important part of all poisonings. Causes such as wrong drug use, wrong dose, drug-drug interactions, drug side effects, abused over-the-counter drugs, and exposure to chemical and biological agents constitute an important part of poisoning. The severity of poisoning may vary depending on the type, amount, and personal characteristics of the toxic substance. Organ involvements such as gastrointestinal, central nervous system, respiratory system, cardiovascular, eye, skin, and mucous membranes are seen[6]. For example, symptoms such as altered consciousness, tachycardia/bradycardia, or hypertension/hypotension may be seen because the cardiovascular system is being affected. Changes in clinical findings and ECG may be revealed according to the degree of heart involvement. It has a wide spectrum that progresses from a completely asymptomatic condition to death. Rapid recognition and effective intervention by the emergency physician are of great importance.

This review considers the use of ECG in the management of poisoned patients.

Cardiovascular system involvement

Intoxications, which are accepted as "reversible causes" in cardiac arrest patients, have an important place in advanced cardiac life support[7]. One of the most common causes of death from poisoning is the clinical picture that occurs because the cardiovascular system is affected. The mortality rate in acute

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poisoning with cardiotoxic drugs is approximately 12%[8]. The cause of death is cardiovascular dysfunction, arrhythmias, and collapses. Emergency physicians may encounter many unforeseen situations when dealing with patients with a drug overdose. Multiple organ toxicity findings may progress to result in a cardiovascular collapse. Therefore, identification of a possible 'toxidrome' is invaluable in the management of poisoned patients. Thus, instead of focusing on the toxins, we will be able to intervene appropriately for the poisoned patient. Meanwhile, an easily taken electrocardiogram of the patient can provide us with valuable information.

Electrocardiography

Electrocardiography is conduction recordings obtained by recording the electrical activity occurring in the heart to examine the state of the heart muscle and conduction network[9]. It is frequently used in emergency conditions and rapid evaluations, especially because it is fast, easily accessible, inexpensive, and non-invasive. With ECG, it can easily evaluate ischemic and structural heart diseases, cardiac involvement of systemic diseases, lung diseases, and electrolyte disorders, and give us an idea about drug side effects and some poisonings. Electrocardiography can be more helpful in poisoning when evaluated together with examination findings, tests, and films. ECG has an important place in the management of poisonings; It is used for imaging, diagnosis, prognosis prediction, and patient monitoring. In poisonings, cardiotoxicity should be well illuminated to make an accurate ECG interpretation. Special needs to know about the mechanisms of cardiotoxicity: sodium channel blockade (Phase 0 prolongation, wide QRS, +R wave in aVR, right axis deviation, ventricular tachycardia and fibrillation, Brugada-like shape). K + channel blockade (prolongation in phases 2 and 3, long QT, Torsades de pointes) (Figure 1).

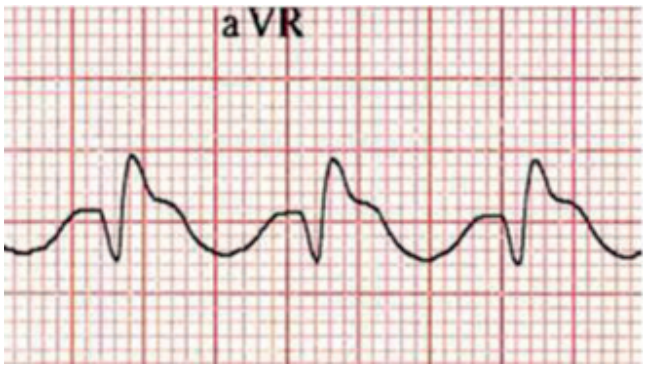

Figure 1: R-wave AVR

Beta-adrenergic receptor blockade (Sinus bradycardia, AV blocks, ventricular bradycardia). Calcium channel blockade (Sinus Bradycardia, 2nd, and 3rd-degree block, asystole)[10]. It is important to have experience in ECG interpretation, first, it is necessary to have an accurate evaluation experience in patients with chest pain, electrolyte disturbance, and shortness of breath. Because an intoxicated person may show ECG changes, which are common causes of coronary artery disease, electrolyte abnormalities, or dyspnea. ECG findings suggest electrolyte disorders such as hypokalemia, hyperkalemia, hypomagnesemia, and hypocalcemia may occur because of poisoning. Therefore, in poisoned patients, it may be useful to check whether there is an old ECG belonging to the patient and to take a serial ECG to the patient. In this way, the effects of delayed-release drugs or late-onset side effects on the heart can be evaluated. One of the most well-known pathophysiological mechanisms is the task of potassium flow from the myocardial membrane through cardiac repolarization. It occurs by blocking the outflow of potassium from the cell in Phase 3, which is the repolarization phase[11]. A possible potassium channel blockade presents as a prolonged QT interval. Also, blocking potassium channels can cause T wave abnormalities or the appearance of U waves. A long QT interval can cause torsades de pointes[12]. Antifungals (fluconazole, itraconazole, ketoconazole), diphenhydramine, amantadine, amiodarone, macrolides, etc. ECG findings include prolongation of the QT interval, sinus tachycardia, torsades de pointes (polymorphic ventricular tachycardia), and ventricular fibrillation[13]. Digoxin is the most used cardiac-positive inotropic agent. It shows its effects by inhibiting the Na-K ATPase pump in the myocardial cell membrane. Digoxin intoxications are frequently encountered due to the narrow therapeutic and toxic dose range of the drug. Ingestion of high doses of digoxin, either by accident or by suicide, can lead to acute poisoning. As the blood digoxin level rises, there is an increase in automaticity in the Purkinje fibers[14]. As a result, ectopic foci become active and ventricular ectopic beats occur. Therefore, it can cause multiple dysrhythmias. These emerging ectopic beats are the first indications that the digoxin concentration has reached the toxic level. Increased automaticity is seen because of increased intracellular calcium. As a result of increased vagal effects in the AV node, suppression of AV conduction is observed. Classic digoxin toxic dysrhythmia: Supraventricular tachycardia (increased automaticity) and slow ventricular response (suppressed AV conduction). Other arrhythmias include frequent premature ventricular complexes, including ventricular bigemia and trigeminal, sinus bradycardia or slow AF, and any type of AV block15. An effective and timely intervention in rhythm disorders, which can be seen in acute digoxin intoxications and can be fatal, can give good results. Downward ST depression inverted or biphasic T waves, and a shortened QT interval are typical with the characteristic "sagging" image. This appearance on the ECG is called the Salvador Dali mustache (Figure 2).

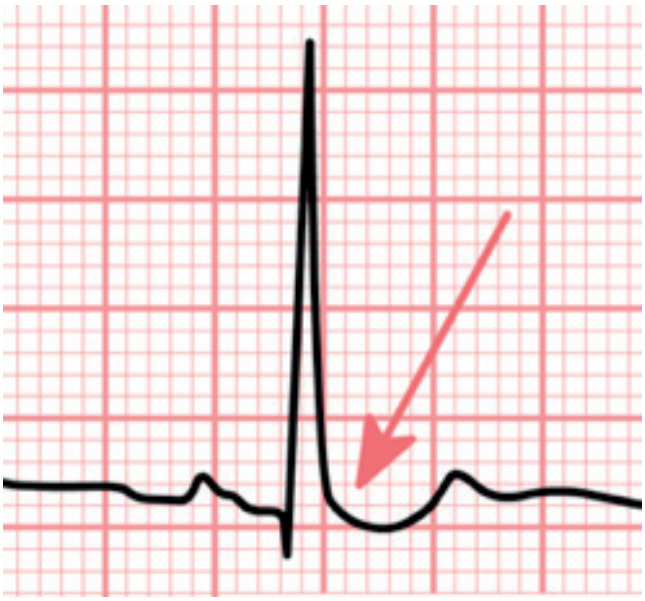

Figure 2: Digoxin intoxications

Beta-adrenergic receptor antagonists are a group of drugs commonly used in the treatment of hypertension and cardiac arrhythmia. Especially propranolol poisonings are more common than others[16]. For rapid recognition of beta-blocker poisoning treatment, we need to know the mechanism of action. With intracellular activation of beta receptors, cyclic adenosine monophosphate increases, protein kinase is activated, and intracellular calcium entry increases. In the blockade of this receptor, the contraction rate of the heart muscle and the AV node conduction rate decrease. Bradycardia and hypotension are the most common findings in beta-blocker poisoning. In overdose, cardiogenic shock may develop due to deep myocardial depression. ECG findings of poisoning include sinus bradycardia and decreased AV nodal conduction (1st to 3rd-degree heart blocks). Special consideration should be given to Propranolol (Na channel block effect) and Sotalol (K+ channel block effect). Propanolol: Acts as a TCA, blocks Na channels so that the QRS widens and a +R wave is seen in aVR. Sotalol: Blocks K channels, thereby prolonging the QT. Torsades de Pointes may develop[17].

Intoxications with tricyclic antidepressants (TCA) mediate their cardiotoxic effects through blockade of myocardial fast sodium channels (QRS prolongation, long R wave in aVR), inhibition of potassium channels (QTc prolongation), and direct myocardial depression[18]. Other toxic effects are produced by the blockade of muscarinic (M1), histamine (H1), and alpha1 adrenergic receptors. The two main side effects of poisoning with a sodium channel blocker are seizures and ventricular rhythm disturbances. Typical ECG of TCA poisoning shows: sinus tachycardia with first-degree AV block (P waves hidden in T waves, best seen in V1-2), wide QRS complexes, and positive R' wave in aVR. Significant QRS broadening, producing a sine wave image resembling hyperkalemia, indicates worsening TCA toxicity. There are situations that may be like ECG findings in TCA poisoning. Because. Poisonings whose mechanism of action is sodium channel blocker are quite common. Tricyclic antidepressants, Type Ia antiarrhythmics (quinidine, procainamide), Type Ic antiarrhythmics (flecainide, encainide), Local anesthetics (bupivacaine, ropivacaine), Antimalarials (chloroquine, hydroxychloroquine), Dextropropoxyphene, Propranolol, Kini[19]. Agents that antagonize calcium channels cause myocardial depression by reducing calcium entry into cells. Bradycardia develops because of calcium channel antagonism in the SA and AV nodes. If peripheral hypotensive effects prevail, reflex tachycardia may occur. Calcium channel blockers such as dihydropyridine often cause sinus bradycardia, and AV nodal conduction blocks (1st to 3rd-degree heart blocks).

Discussion

Evaluation of electrocardiography

Systematic evaluation of the ECG in a patient followed up with poisoning is essential for details that may be overlooked. Velocity, rhythm, intervals, and segments, QRS, wave morphologies, durations, and ischemic changes should be followed carefully[20]. In unstable patients, clues to drug cardiotoxicity should be sought when performing rhythm analysis. Are there ectopic beats on the ECG? The answer to this question may carry important clues. Automaticity caused by sympathomimetics may underlie ectopic beats. This may be the first sign of a problem caused by acute coronary syndrome or electrolyte disturbances. Is the rhythm supraventricular? or ventricular? Is bradycardia with AV block? or without AV block? Is tachycardia narrow complex? Or is it a large complex? Answers to questions such as: For life-threatening rhythms, ventricular tachycardia, ventricular fibrillation, and complete AV-block, the guidelines developed should be followed and first intervention should be made[21].

Agents that can cause tachycardia; are sympathomimetics (methamphetamine), anticholinergics (antidepressants, antipsychotics), class 1A and 1C antidysrhythmics, and TCA[22]. Agents that can cause bradycardia; calcium channel / beta blockers / digoxin (AV block), opioids / ethanol, organophosphates, lithium. Prolonging the PR interval may indicate beta-adrenergic antagonism, calcium channel antagonism, or digoxin poisoning[23]. Also, opioids via sympathomimetics and vagal tone. Clonidine or sedative hypnotics may be responsible for this effect. Examination of the PR interval is vitally important for the evaluation of AV blocks. A prolonged QRS suggests sodium channel blockade. TCAs, Class 1A antidysrhythmics, carbamazepine, right branch primarily blocked, obvious R wave – aVR. It should be noted that there is a list of drugs that prolong the QRS duration. The right-sided intraventricular conduction system is more sensitive to the toxic effects of some sodium channel blockers than the left bundle. Delayed depolarization of the

right ventricle causes changes in the morphology of the QRS complex in aVR. Therefore, there are prominent R waves in the AVR. In addition, it should not be forgotten that the Brugada pattern is among the other signs of right ventricular depolarization delay. J wave or Osborn wave in patients with hypothermia[24]. Visible. It is possible to occur due to syncope and prolonged exposure to cold after an overdose of medication. A hypothermic patient has bradycardia on the ECG strip and a positive deviation at the QRS/ST junction known as an Osborn or J wave (marked with an arrow). ST changes are closely associated with ischemia. Cardiac evaluation and follow-up are very important in reducing mortality in CO poisonings[25]. Although myocardial ischemia is an expected clinical condition in carbon monoxide poisoning, ST segment elevation can be seen in the ECG. Coronary anatomy may appear completely normal in the angiograms of these patients. If drug overdose, such as cocaine, has triggered a vasoconstriction, it may produce ST segment depressions or elevations suggestive of ischemia. Cyanide poisoning, which causes severe ischemia in tissues, and hypotension caused by calcium channel blockers may also present with ST changes. Brugada pattern can be described as a kind of ST segment elevation. Again, ST segment changes may be caused by digoxin poisoning. The QT interval may be prolonged in overdose of drugs with cardiotoxicity. In addition, although due to potassium channel blockade, prolonged QRS due to sodium or calcium channel antagonism may also increase the QT interval. Just like the list of drugs that affect the QRS duration, there is a long list of drugs that prolong the QT duration. In toxicology, absolute QT is a better predictor of Torsades risk than corrected QT (QTc). Therefore, the QT nomogram is best for risk estimation. Formulated QT corrected for QT duration, which can vary with heart rate. In general, a QTc greater than 450 ms in men and 470 ms in women is considered prolonged. Poisoned patients should be evaluated in a lead with good T waves (Figure 3).

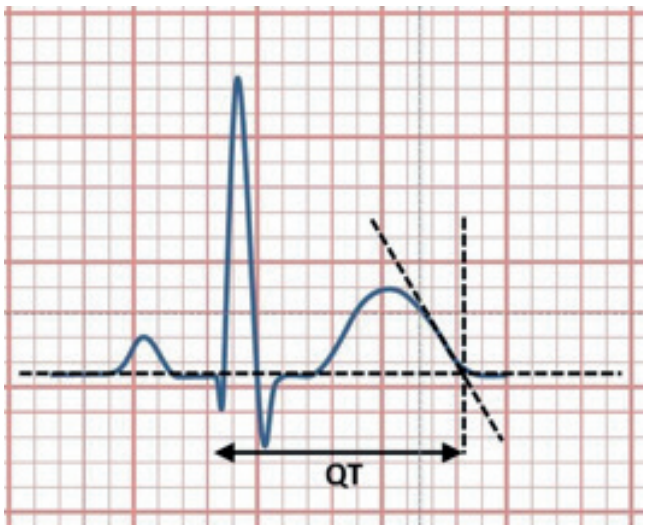

Figure 3: QT interval

It should be noted that QTc prolongation is associated with cardiac death[26]. Mad honey poisoning occurs by consuming honey obtained from plants belonging to the Rhododendron Ponticum and Luteum family, which are often grown in the Eastern Black Sea region in our country[27]. Electrocardiography has a critical importance in the diagnosis. Complication rates can be easily reduced with early diagnosis and effective treatment. In cases of poisoning caused by sodium channel blockade; Changes were observed in QRS duration, QT interval length, and atrial-ventricular depolarization-repolarization parameters[28]. ECG changes seen with symptoms and physical findings in cases of overdose are called "ECG toxidromes". ECG findings that should especially be sought in poisoned patients presenting with sinus rhythm; Signs of AV block, long QTc, or sodium channel blockade (R in aVR). In poisoned patients presenting with bradycardia, attention should be paid to blood glucose monitoring, AV blocks, and prolonged QT. Bradycardia and long QT opioids, bradycardia and nystagmus lithium, bradycardia and AV block may suggest calcium channel blocker poisoning and hyperglycemia, and bradycardia and cholinergic symptoms may suggest organophosphate poisoning. Tachycardia is a common presentation rhythm in poisoned patients. ECG of tachycardic patients suggests wide QRS complexes and sodium channel blockade (R wave in aVR). Wide QRS with tachycardia may suggest TCA, diphenhydramine, phenothiazine, bupropion, and cocaine overdoses[29]. A narrow QRS suggests antidepressants and antipsychotics, hypovolemia, acetaminophen, and carbon monoxide poisoning. In case of poisoning and its approach to ECG; rate and rhythm assessment, PR interval – looking for any degree of heart block, and II. Evaluating the QRS duration in the lead is the most important part[30].

Conclusion

In patients presenting with poisoning, the ECG should be taken and evaluated within the first 10 minutes. If the poisoning agent is an agent that influences the cardiovascular system, it should be kept in mind that continuous cardiac monitoring and control ECG evaluation should be performed in addition to the application of ECG. While evaluating the ECG, the duration of the QRS in DII should be specially checked.

References

1. Morrison EE, Sandilands EA. Principles of management of the poisoned patient. Medicine. 2020; 48(3): 160–4.
2. Hovda LR, Brutlag AG, Poppenga RH, Peterson KL. Emergency Management of the Poisoned Patient. Small Anim Toxicol. 2016: 19.
3. Kekec Z, Sozuer EM, Duymaz H, Okkan S. Evaluatian of the patients applied to the emergency department due to multiple

drug poisoning: analysis of 7 years. Turkish Journal of Emergency Medicine 2005; 5: 69-72.

4. Chelkeba L, Mulatu A, Feyissa D, et al. Patterns and epidemiology of acute poisoning in Ethiopia: systematic review of observational studies. Arch Public Health. 2018; 76(1): 34.
5. Dart RC, Bronstein AC, Spyker DA, et al. Poisoning in the United States: 2012 emergency medicine report of the National Poison Data System. Ann Emerg Med. 2015; 65: 416-22.
6. Bai L, Peng X, Liu Y, Sun Y, Zheng L, Liu Z, Wan K, Wang J, Zhao J, Qiu Z. Association between acute severe mercury poisoning and multiple organ failure. Am J Transl Res. 2020; 12(8): 4347-433.
7. Lott C, Alfonzo A, Barelli A, et al. European Resuscitation Council Guidelines 2021: cardiac arrest in special circumstances. Resuscitation 2021; 161.
8. Johnson NH, Gaieski DF, Allen SR, et al. A review of emergency cardiopulmonary bypass for severe poisoning by cardiotoxic drugs. J Med Toxicol. 2013;99(1):54–60.
9. Mirvis MGL (2001) Electrocardiography. In: Braunwald E, Zipes DP, Peter L (eds) Heart disease. Saunders, Philadelphia, pp 82–125.
10. Seger DL. A critical reconsideration of the clinical effects and treatment recommendations for sodium channel blocking drug cardiotoxicity. Toxicol. Rev. 2006; 25: 283–296.
11. Ponte ML, Keller GA, Di Girolamo G. Mechanisms of drug induced QT interval prolongation. Curr Drug Saf. 2010;5:44–53.
12. Klein MG, Krantz MJ, Fatima N, Watters A, Colon-Sanchez D, Geiger RM, Goldstein RE, Solhjoo S, Mehler PS, Flagg TP, et al. Methadone blockade of cardiac inward rectifier K+ current augments membrane instability and amplifies U waves on surface ECGs: a translational study. J Am Heart Assoc. 2022; 11:e023482.
13. Han J, Ackerman MJ, Moir C, Cai C, Xiao P-L, Zhang P et al. Left cardiac sympathetic denervation reduces skin sympathetic nerve activity in patients with long QT syndrome. Heart Rhythm. 2020;17(10): 1639-45.
14. Lind L, Araujo JA, Barehowsky A, Belcher S, Berridge BR, Chiamvimonvat N., et al. (2021). Key characteristics of cardiovascular toxicants. Environmental Health Perspectives, 129(9), 95001–95001.
15. Sheldon SH, Gard JJ, Asirvatham SJ. Premature ventricular contractions and non-sustained ventricular tachycardia: association with sudden cardiac death, risk stratification, and management strategies. Indian Pacing Electrophysiol J. 2010; 10:357–371.
16. Reith DM, Dawson AH, Epid D, et al. Relative toxicity of beta blockers in overdose. J Toxicol Clin Toxicol 1996; 34(3): 273–8.
17. Hennersdorf MG, Strauer BE. Arterial hypertension and cardiac arrhythmias. J Hypertens. 2001; 19: 167–177.
18. M.X. Hu, Y. Milaneschi, F. Lamers, I.M. Nolte, H. Snieder, C.V. Dolan, B. Penninx, E.J.C. de Geus. The association of depression and anxiety with cardiac autonomic activity: the role of confounding effects of antidepressants Depress. Anxiety, 36 (2019), pp. 1163-72.
19. Giwa A, Oey E (2018) The return of an old nemesis: survival after severe tricyclic antidepressant toxicity, a case report. Toxicol Rep 10(5):357–362.
20. Bhoi AK, Sherpa KS. Statistical analysis of QRS-complex to evaluate the QR versus RS interval alteration during ischemia. J. Med. Imag. Health Inform. 6(1), 210–214.
21. Acharya UR, Fujita H, Oh SL, Raghavendra U, Tan JH, Adam M, Gertych A, Hagiwara Y. Automated identification of shockable and non-shockable life-threatening ventricular arrhythmias usingconvolutional neural network. Future Gener. Comput. Syst.2018,79, 952–959.
22. Mladěnka P, Patočka LAJ et al., "Comprehensive review of cardiovascular toxicity of drugs and related agents," Medicinal Research Reviews, vol. 38, pp. 2018; 1332–1403.
23. Chen X, Peng YW, Han X, Liu Y, Lin XC, Cui Y. Sixteen isostructural phosphonate metal-organic frameworks with controlled Lewis acidity and chemical stability for asymmetric catalysis. Nat. Commun.2017, 8, 2171.
24. Antzelevitch C. Molecular biology and cellular mechanisms of Brugada and long QT syndromes in infants and young children. J Electrocardiol. 2001; 34: 177-181.
25. Lippi G, Rastelli G, Meschi T, Borghi L, Cervellin G. Pathophysiology, clinics, diagnosis and treatment of heart involvement in carbon monoxide poisoning. Clin Biochem 2012;45:1278–1285.
26. Hoffman, R.S. Treatment of patients with cocaine-induced arrhythmias: Bringing the bench to the bedside. Br. J. Clin. Pharmacol.2010,69, 448–457.
27. Jansen SA, Kleerekooper I, Hofman ZL, Kappen IF, Stary-Weinzinger A, van der Heyden MA. Grayanotoxin poisoning: 'mad honey disease' and beyond. Cardiovasc Toxicol. 2012;12:208–15.
28. De Bie J, Diemberger I, Mason JW. Comparison of PR, QRS, and QT interval measurements by seven ECG interpretation programs. J Electrocardiol. 2020;63:75–82.
29. Yates, C, Manini, FA. Utility of the electrocardiogram in drug overdose and poisoning: theoretical considerations and clinical implications. Curr Cardiol Rev 2012; 8: 137–151.
30. Wills B, Theeler BJ, Ney JP. Drug- and toxin-associated seizures. In: Dobbs MR, editor. Clinical neurotoxicology: syndromes, substances, environments. Philadelphia: Saunders Elsevier; 2009. p. 131–50.

Case Report

Eurasian Journal of Toxicology

A Case Report: Datura Intoxication in The Emergency Department

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Abstract

Datura Stramonium (DS) is a in our country; it grows wild in rural areas without the need for agriculture. DS can also be used as herbal medicine in respiratory system diseases such as asthma and bronchitis, in hemorrhoids and dermatological diseases such as eczema. It may lead to anticholinergic symptoms such as mydriasis, tachycardia, dry skin, flushing and urinary retention in humans due to its atropine content, and when it affects the central nervous system, it may cause symptoms ranging from restlessness, agitation, hallucination, convulsion to coma and death. We aimed to describe the case of the patient who was taken to the emergency department with the pre-diagnosis of altered consciousness, agitation, and psychiatric disorder, progressed with the signs of delirium and anticholinergic toxicity, was diagnosed with Datura Stramonium intoxication, was admitted to the intensive care unit after the first treatment, and was discharged with recovery.

Keywords: Datura Stramonium, Intoxication, Emergency Department.

Introduction

Datura Stramonium is a foul-smelling plant with a height of 60 to 150 cm, which belongs to the Solanaceae grass family, gives an average of 8-10 branches, and has white funnel-shaped flowers (the reason why it is called "tube flower" is because of the appearance of its flowers), green fruits of 3-5 cm in size, leaves varying between 8 and 20 cm, and an ability to regenerate annually[1]. It grows naturally in rural areas of our country without needing any farming. It is locally called by different names such as thorn apple, jimsonweed, tube flower, mad apple, jimson, fireweed, devil's apple, jasmine, apple of peru, and eggplant herb[2].

The main medicinal value of the plant is its alkaloid content. The majority of these alkaloids consist of tropane ring containing L-hyoscyamine, and atropine formed by racemization from scopolamine and L-hyoscyamine.

Datura Stramonium can also be used as herbal medicine in respiratory system diseases such as asthma and bronchitis, in hemorrhoids and dermatological diseases such as eczema[3]. It may lead to anticholinergic symptoms such as mydriasis, tachycardia, dry skin, flushing and urinary retention in humans due to its atropine content, and when it affects the central nervous system, it may cause delirium symptoms ranging from restlessness, agitation, hallucination, convulsion to coma and death[4]. Delirium is defined as a neuropsychiatric syndrome which is frequently encountered in the emergency department[5,6].

We aimed to present the patient who was admitted to the emergency department with the pre-diagnosis of altered consciousness, agitation, and psychiatric disorder, had the symptoms of delirium and anticholinergic toxicity, and then who was diagnosed with Datura Stramonium intoxication, hospitalized in the intensive care unit after the first treatment and discharged with recovery.

Case

A 40-year-old male patient, a pump attendant in the fuel station in a rural area, was brought to our emergency department by his colleagues with complaints of agitation, restlessness, altered consciousness, irritability, and harming himself and those around him. During admission, his general condition was

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moderate, his orientation and cooperation were impaired, and he was restless and agitated, was talking to himself senselessly, and had hallucinogenic attitudes. When the patient's vital signs were examined, his arterial blood pressure was 150/90 mmHg, his pulse rate was 120/min, and he had a body temperature of 37°C. He had pupillary mydriasis, blurred vision, dry skin and mouth, tachycardic heart rhythm, decreased bowel sounds in the abdomen, no defense or rebound.

The patient's biochemistry tests (Glucose, BUN, Creatinine, AST, ALT, Amylase, Lipase, Sodium, Potassium, Chlorine), complete blood count, blood gas and carboxyhemoglobin (CoHb) levels examined in the emergency department laboratory were found to be normal. No acute pathology was detected in the cranial tomography of the patient. Electrocardiography revealed no finding other than sinus tachycardia.

In the anamnesis taken from the patient's relatives, the presence of sudden onset of symptoms, the patient's interest in herbal products, and the symptoms consistent with anticholinergic syndrome suggested that he could be a case of acute intoxication. When the patient's relatives were asked for the plant he ate at his workplace, it was learned that the plant brought was Datura Stramonium (Photos 1 and 2). The patient was given a benzodiazepine for sedation in the emergency department, he was hydrated, a urinary catheter was inserted for possible urinary obstruction, and a nasogastric tube was inserted for possible ileus. The patient was hospitalized in the anesthesia intensive care unit for follow-up. The patient's symptoms regressed and disappeared within 24 hours. He was discharged with full recovery. When the patient was contacted later, it was learned that he ate only a few seeds of the plant for the treatment of diarrhea.

Discussion

Datura stramonium is a plant known by the names of "jimsonweed, jimpson, tube flower, bellflower, wild bishop, convolvulus sepium, eggplant flower, jimson, apple of peru, magic grass, devil's apple". It is usually used unconsciously as a medicine. It is commonly used among people by considering

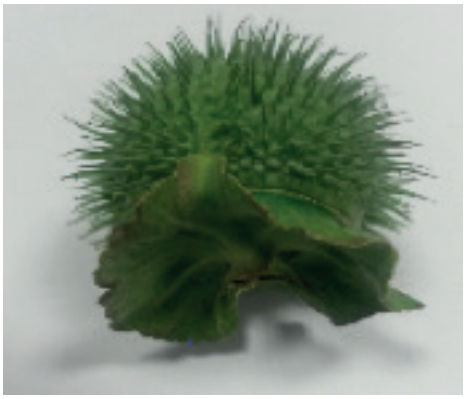

Photo 1: Photos were taken by us, All rights are reserved by us.

Photo 2: Photos were taken by us, All rights are reserved by us.

that it is useful in asthma, diarrhea, and gastrointestinal problems. Furthermore, it is also used as a narcotic drug[7]. Our patient used it for the treatment of diarrhea. In case of high doses, it may lead to tachycardia, mydriasis, flushing, restlessness, perceptual disorders and agitation caused by atropine. The symptoms begin approximately 30 minutes after oral intake[8,9]. Our patient had complaints of agitation, restlessness, altered consciousness, irritability, and harming himself and those around him. Anticholinergic syndrome can be caused by a group of drugs or substances that prevent acetylcholine from binding to muscarinic receptors[10,11]. Drug-related causes include antipsychotics, tricyclic antidepressants (TSA), antihistamines, carbamazepine, atropine and drugs containing scopolamine[12]. Furthermore, Datura stramonium and belladonnae (Herba Belladonnae), the leaves of which are consumed as tea in our country and around the world and that are used uncontrollably in the treatment of gastrointestinal problems, hemorrhoids, asthma and bronchitis through cigarettes prepared from the leaves due to their antispasmodic effects, are also among the causes of anticholinergic syndrome. The symptoms such as blurred vision, tachycardia, inability to urinate, and mouth dryness, and ileus, facial flushing, dysrhythmia, auditory and visual hallucinations, and convulsions can also be detected after using it[13]. Indeed, our patient also had anticholinergic symptoms and signs such as tachycardia, mydriasis, restlessness, hallucinations, and dry mouth.

In patients with suspected anticholinergic toxicity, necessary antidote treatment should be administered after circulation, airway and respiration are secured[10]. Since the patient did not have a problem of airway protection when he presented to us, no intervention such as intubation was required. Furthermore, conservative treatment is generally performed in anticholinergic toxicity. Its specific antidote is 'physostigmine'. Physostigmine reversibly inhibits anticholinesterase and is also effective in neurological symptoms since it crosses the blood-brain barrier. Most of the cases can be treated without administering physostigmine. Physostigmine is recommended to be used if tachycardia, coma and convulsion conditions causing hemodynamic instability, and severe respiratory depression are present10. The current findings were not detected in our patient, so

there was no need to administer physostigmine. The use of physostigmine in cases without signs of anticholinergic toxicity leads to cholinergic symptoms such as bronchospasm, bronchorea, convulsions, and bradyarrhythmia[11]. If the patient is very agitated, benzodiazepines can be used for sedation. In our patient, sedation was performed with benzodiazepines since agitation was high.

Furthermore, poisoning was caused by consuming a large number of plant seeds in cases in the literature, however, unlike the literature, our patient ate a few seeds. Therefore, it is necessary to consider that anticholinergic symptoms can also be observed due to very little consumption.

Conclusion

There are many admissions to the emergency department due to agitation and altered consciousness. Although cerebrovascular and psychiatric diseases first come to mind in the preliminary diagnosis of these patients, delirium (organic brain syndrome) should be evaluated in the differential diagnosis, and intoxications should definitely be considered in sudden-onset cases. Furthermore, although drug-related poisoning is first considered in cases of intoxication, plant intoxications should also be kept in mind and examined in detail in the anamnesis due to alternative medicine and phytotherapy methods, which are increasingly used today.

References

1. Kurt DD, Korulmaz A, Alakaya M, Arslanköylü AE. Deliryuma yol açan nadir bir intoksikasyon: Datura stramonium. Mersin Üniversitesi Sağlık Bilimleri Dergisi. 2022; 15(1): 128-132.
2. Taştekin F, Işikli N. Antikolinerjik İntoksikasyonlu Bir Vaka: Datura Stramonium Zehirlenmesi. Turkiye Klinikleri J Intern Med. 2020;5(1):42-5.
3. Işıkay S. Datura Stramonium Zehirlenmesi: Bir Vaka Sunumu. AKATOS 2011; 2:26-28.
4. Disel NR, Yilmaz M, Kekeç Z. Karanlık M Poisoned after dinner: Dolma with Datura stramonium. Turk J Emerg Med 2016;15;51-55.
5. Mattoo SK, Grover S, Gupta N. Delirium in general practice. Indian J Med Res 2010; 131:387-398.
6. Guenther U, et al. "Validity and reliability of the CAM-ICU Flowsheet to diagnose delirium in surgical ICU patients." Journal of critical care. 2010; 25(1): 144-151.
7. Köse R, Şahin ÖÖ, Abay E. Datura stramonium zehirlenmesine bağlı deliryum: Bir olgu. Anadolu Psikiyatri Dergisi 2008; 9:54-57.
8. Gaire BP. and Subedi L. A review on the pharmacological and toxicological aspects of Datura stramonium L. Journal of integrative medicine 2013; 11(2): 73-79.
9. Ram A, et al. Medicinal plants useful for treating chronic obstructive pulmonary disease (COPD): current status and future perspectives. Fitoterapia 2011 82(2): 141-151.
10. Krenzelok EP. Aspects of Datura poisoning and treatment. Clinical Toxicology 2010 48 (2): 104-110.
11. Dündar D. Fizostigmin Salisilat. Editörler; Satar S, İkizceli İ. Goldfrank'ın Toksikolojik Aciller El Kitabı. Adana, Nobel Tıp Kitabevi.2008; 439-440.
12. Dede G, Semra Karaçayır S. Datura stramonium intoxication: a case report. Ege Klin Tıp Derg. 2017;55(1):36-8.
13. Karadaş S, Selvi Y, *Şahin* M, Selvi F, *Öncü* R, *Özgökçe* F. Datura stramonium zehirlenmesi: Psikiyatrik belirtilerle başvuran olgu. Düşünen Adam: Psikiyatri ve Nörolojik Bilimler Dergisi: 2011;24:152-154.