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Venomous Snakebites in Southern Croatia

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A B S T R A C T

*This retrospective study is based on the analysis of 542 snakebite envenomation cases in southern Croatia, which were treated in the University Hospital Split over the period of 21 years. The aim of this study was to determine the incidence of venomous snakebite in southern Croatia, epidemiological and clinical features of snakebite and treatment in the region. The mean annual snakebite incidence in southern Croatia was 5.2 per 100,000 inhabitants. The nose- horned viper (*Vipera ammodytes*) was responsible for most bites, only a small proportion being inflicted by the adder (*Vipera berus*). People of all ages were affected (1 – 82 year old), but the bites were more frequent in individuals older than 50 (46% of the cases) and in children and adolescents 19 year old and younger (27% of the cases). Most snakebite accidents happened in warm spring and summer months, the highest number occurring in May (22%). A majority of the victims were rural people engaged in agricultural activities. Bites on the upper limbs were more frequent (57%) than bites on the lower limbs (42%). With regard to envenomation severity, there were 15.1% minor, 40.5% mild, 26% moderate and 18% severe cases. Two victims died (0.4%). The antivenom produced by the Institute of Immunology in Zagreb was given to virtually all patients, and complications following its administration were rare. The antivenom was used more often than it was suggested by the symptoms present.*

Key words: *snakebite, snake venom, viper venom, antivenom, epidemiology, Croatia*

Introduction

Venomous snakebites are common in tropical and subtropical regions, but they also occur in regions with temperate climates[1–4]. The only venomous snakes in Europe are viperids (belonging to the family *Viperidae*), specifically »true« vipers (*Viperinae* subfamily)[5]. Croatia is situated in a region of Europe where vipers are abundant. Europe's most venomous snake, the nose-horned viper, *Vipera ammodytes*, is frequently encountered in dry, rocky areas of southern Croatia[6]. Most of its victims are rural people engaged in agricultural activities. *V. ammodytes* delivers on average 20 mg of dry venom in a single bite, which is considered to be a lethal dose for humans. The bites of the adder (*V. berus*) in Croatia are much less frequent[5]. The venoms of *V. ammodytes* and *V. berus*, as well as the venoms of other *Viperinae*, are largely necrotising and hemorrhagic, and only to a lesser degree neurotoxic. These venoms contain active protein substances (enzymes and toxins) with different mechanisms of activity[5,7]. At the time of envenomation a potent inoculum of aerobic and anaerobic bacteria is introduced by the snake's fangs. These bacteria can cause serious infections in necrotic tissue at the bite site[4,5]. Most studies carried out so far in Europe deal with snakebites caused by *V. berus* and *V. aspis*[1,3,8,9]. Hence, it was very interesting to conduct research in a region where the bites are inflicted primarily by *V. ammodytes*.

In this paper, we present the incidence of venomous snakebite, epidemiological trends, clinical manifestation, and snakebite treatment in southern Croatia.

Patients and Methods

This study is retrospective, and the source of data is the medical records of the snakebite patients that received treatment at the University Hospital of Split (UH) (southern Croatia) over the period of 21 years (from January 1, 1982, to December 31, 2002). The study covered Split-Dalmatia County in southern Croatia, which is inhabited by 496,395 people (according to 1991 census)[10]. By convention, almost all snakebite cases in this region are sent to the Department of Infectious Diseases at the UH Split. After a snakebite victim is brought to the department, he or she is examined by an infectious disease

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specialist, who determines whether or not the venom was injected. Only venomous bite cases are hospitalized. There were likely some mild cases of snakebite envenomation which were not registered, because the patients did not visit the hospital. In most cases, a physician identified the snake involved based on the data given by the patients or their companions. Therefore, it was possible that in some cases the responsible species was misidentified. In certain cases the physician identified the snake involved by observing the killed snake brought to the hospital. The data gathered from the medical records of the patients were assigned to demographic, epidemiological, clinical, laboratory and treatment categories. Zagreb antivenom was the sole antivenom used in treatment. The data were analyzed in Statistica 6.0 (StatSoft, Inc., Tulsa, OK, USA). A portion of the snakebite data from the medical records of the UH Split were also used in another study by Radonić et al., 1997[7].

The mean annual snakebite incidence in southern Croatia was obtained in the following way. For a particular year, the incidence was calculated by dividing the number of snakebite victims treated at the hospital with the number of residents of the region, and then by multiplying this result by a constant (100,000). The incidence values for all years were added and the sum obtained was divided by 21 (the study period of 21 years), which gave the mean annual snakebite incidence per 100,000 inhabitants.

For the classification of the nose-horned viper and adder bite reaction, we used the scheme devised by Reid (1976) and modified by Persson (1981):

- Minor reaction-local oedema, no general symptoms and signs except those of fright.
- Mild reaction-local or more extensive oedema, with or without gastrointestinal signs and symptoms but without other systemic effects.
- Moderate reaction-extensive oedema, shock lasting for less than two hours, other signs and symptoms of moderate poisoning.
- Severe reaction-shock lasting for more than two hours or recurring episodes of shock, other signs and symptoms of severe systemic poisoning.
- Fatal reaction-more evident systemic poisoning ending in death of the patients[1,11,12].

Results

Epidemiological data

The mean annual snakebite incidence in southern Croatia was 5.2 per 100,000 inhabitants. This snakebite incidence would have likely been higher if the mild envenomation cases which did not visit the hospital had been documented. Two snakebite deaths were recorded (0.4%). In one case, the individual was bitten in the neck while sleeping in a sleeping bag. The other individual was bitten in the armpit region while resting with the hand put on a dry-stone wall. It is likely that the venom was injected directly into a blood vessel, which would lead to toxic shock. It is important to point out that these two fatalities occurred during military operations in 1993, and that the victims did not receive adequate medical treatment[7]. The bites of *V. ammodytes* were much more frequent than the bites of *V. berus*. In 9% of the snakebite cases we could positively identify the species based on the dead specimen brought to the hospital, and in these cases the ratio of *V. ammodytes* to *V. berus* specimens was 16:1. In the remaining cases, 11% of them did not see the snake at all, 45% saw the snake, but did not recognize it, 33% believe they were bitten by *V. ammodytes*, and 2% believe to be bitten by *V. berus*. People of all ages suffered bites (1 to 82 years of age), but the bites were more frequent in individuals older than 50 (252 cases or 46%) and in children and adolescents 19 year old and younger (148 cases or 29%) (Figure 1). We found the following professions or groups of people to be especially vulnerable to snakebites. At highest risk were the rural people engaged in agricultural activities (planting, harvesting, etc), comprising 45% of the cases. Other vulnerable groups were cattle-breeders (10%), people gathering wood (7%) and those gathering wild edible plants such as blackberries and asparagus (5%). The children were bitten while playing outdoors (12%). The remaining 21% of the cases included tourists, backpackers, mountain climbers, hunters, soldiers and individuals who keep venomous snakes

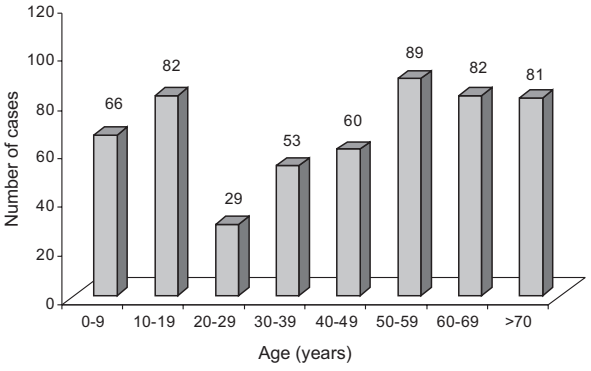

Fig. 1. Age distribution of 542 snakebites envenomation cases in southern Croatia.

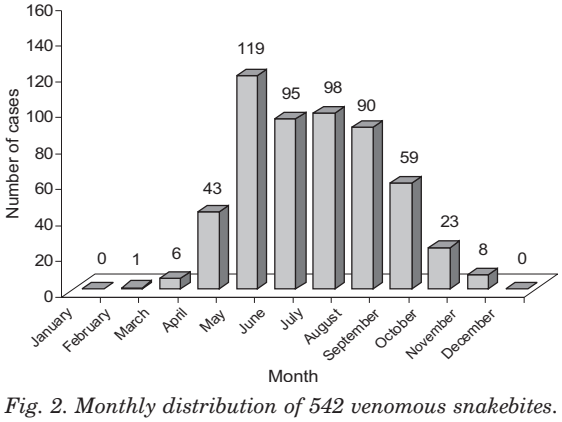

Fig. 2. Monthly distribution of 542 venomous snakebites.

as pets. There was an almost equal number of men and women affected by snakebites (51% and 49%, respectively). Most bites occurred during warm spring and summer months (504 cases or 93%), the most hazardous month being May (119 cases or 22%) (Figure 2). During colder months (February, March, October and November) snakebites were very rare (1–23 cases or 0.2–4.2%). In December and January no snakebite accidents occurred (Figure 2). Bites on the upper limb were more frequent (57%) than bites on the lower limb (42%). The bites on the head, neck, armpit and gluttonous region occurred rarely, a single victim in each case.

Clinical data

Symptoms and signs of *V. ammodytes* and *V. berus* bites are reported in Table 1. Swelling at the bite site was recorded in all patients. The extent of swelling varied from barely noticeable to massive, which spread rapidly over the entire limb and also affected the trunk (Figure 3). Extensive oedema was present in 46.8% of the patients. The extent of swelling was estimated based on the observation of the affected area. Discoloration of skin and ecchymosis were recorded in 92.2% of patients, also varying in extent from minor and barely noticeable to extensive, affecting the entire limb and also the trunk (Figure 3). Regional lymphadenitis was present in 45% of the patients.

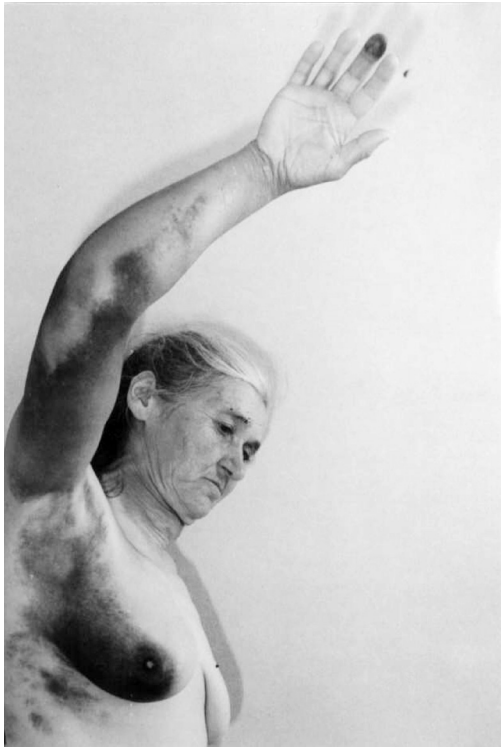

Fig. 3. Vipera ammodytes bite on the right hand finger in a 78-year-old female. Oedema, discoloration of skin and ecchymosis of the whole arm and a part of the body.

TABLE 1
SYMPTOMS AND SIGNS OF ENVENOMATION IN 542 PATIENTS

Symptoms and signs	Number of cases	%
Oedema	542	100.0
Local	288	53.1
Extensive	254	46.8
Skin discoloration and ecchymosis	500	92.2
Regional lymphadenitis	244	45.0
Hemorrhagic blisters	71	13.0
Skin and muscle necrosis	21	3.8
Infection (cellulitis, apscess)	5	0.9
Thrombophlebitis	11	2.0
Compartment syndrome	10	1.8
Gastrointestinal symptoms	122	22.5
Shock	28	5.1
<2 hours	17	3.1
>2 hours	11	2.0
Central nervous system depression	32	5.9
Slight	27	5.0
Unconsciousness	5	0.9
Cranial nerve paresis/paralysis	87	16.0
Fever	330	60.8
Liver damage	24	4.4
Renal failure	25	4.6
Leucocytoses	196	36.1
Anemia	35	6.4
Thrombocytopenia	11	2.0
Prolonged prothrobin time	12	2.2
Hematuria	48	8.8
Melena	7	1.2
Hematemesis	4	0.7
Epistaxis	5	0.9

Other local effects included hemorrhagic blisters (13% of the cases), skin and muscle necrosis (3.8%), thrombophlebitis (2%), compartment syndrome (CS) (1.8%) and infection (cellulitis or abscess) (0.9%). Different sizes of hemorrhagic blisters were observed, from the size of a pea to the size of a walnut and even larger (Figure 4). In most cases, the incision of a hemorrhagic blister was performed. Early debridment was carried out only when the skin and muscles necrosis were extensive. Thrombophlebitis of the blood vessels near the bite site occurred more often in the elderly and more often in cases in which the bite was on the leg. The diagnosis of CS was based on the observation of the clinical signs and symptoms of the syndrome, since the instrument for measuring compartment pressure was not available. Of the 10 (1.8%) CS cases, there were two cases of small children in shock in which the diagnosis of CS was controversial. CS occurred mostly in children, and upper extremities were

Fig. 4. A large hemorrhagic blister at the bite site in a 58-year-old woman bitten in the right hand finger.

Fig. 6. Paresis of the oculomotor nerve (palpabral ptosis) in a 4-year-old child.

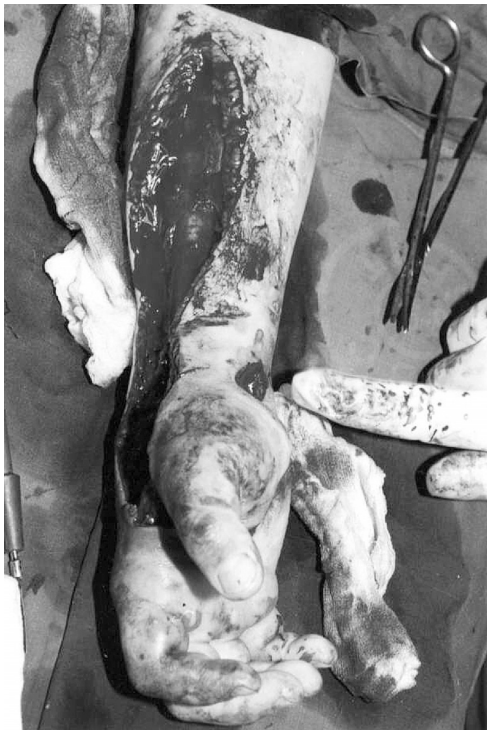

Fig. 5. Dorsal fasciotomy of the forearm and the hand 6 hours after Vipera ammodytes *bite in a 12-year-old boy.*

more affected than lower extremities. Fasciotomies were performed for all CS diagnosed (Figure 5).

Gastrointestinal symptoms (vomiting and diarrhea) were the most common systemic effects, occurring in 22.5% of the patients.

The neurotoxic component of the venom is the cause of the cranial nerve paresis or paralysis in 16% of the cases. The most frequent was the paresis or paralysis of the oculomotor nerve, which was manifested by palpabral ptosis (Figure 6). The paresis or paralysis of the abducens, glossopharyngeal, and other cerebral nerves**,** which were manifested by ophthalmoplegia, dysphagia and dysphonia, were less frequent.

Various degrees of central nervous system (CNS) depression were recorded in 32 (5.9%) cases, and 20 of these cases had shock and hypotension. The remaining 12 CNS depression cases did not have hypotension. Of these 12 cases, 6 had CNS depression only, and 6 had both CNS depression and cranial nerve paralysis or paresis.

Shock occurred in 5.1% of the patients, 2% of whom had hypotension for more than two hours.

Leucocytosis (white blood cell count $>10 \times 10^9$/L) was present in 36.1% of the patients, 10% of the patients having a white blood cell count of more than 15×10^9/L. Anemia was present in 6.4%, thrombocytopenia in 2% and prolonged prothrombin time in 2.2% of the patients. Bleeding also occurred, mostly from the urinary tract (8.8%), and less frequently from the intestinal (1.9%) and respiratory tracts (0.9%). Bleeding was usually mild, mostly not requiring blood transfusions.

Renal failure (oliguria, proteinuria, elevated serum creatinine) was recorded in 4.6% of the patients, and mostly occurred in patients with shock. Liver damage (increased bilirubin, aspartate aminotransferase, alanine aminotransferase) was recorded in 4.4% of the patients. Renal failure and liver damage were temporary, with most patients having recovered by the time they left the hospital.

The body temperature was elevated in 60.8% of the patients, but in most cases it did not exceed 38 °C. Only 5 patients (0.9%) had fever that exceeded 38 °C.

With regard to the severity of envenomation there were 15.1% minor, 40.5% mild, 26% moderate and 18% severe cases. The average duration of hospitalization was 2.6, 5.8, 8.9 and 10.9 days for these four groups, respectively. Two victims died (0.4%) (Table 2).

TABLE 2
DISTRIBUTION OF THE 542 PATIENTS BY SEVERITY OF ENVENOMATION

Reaction	Age group (years)				Total	Days in hospital (average)
	0–9	10–19	20–59	> 60	Number (%)	
Minor	4	12	43	23	82 (15.1)	2.6
Mild	32	36	92	60	220 (40.5)	5.8
Moderate	17	21	51	51	140 (26)	8.9
Severe	13	13	43	29	98 (18)	10.9
Fatal	0	0	2	0	2 (0.4)	0

Treatment

Approximately one-fourth of the patients used first aid, specifically, a constricting band (16.4% of the patients), incision (6%), suction at the bite site (4.6%), and immobilization (2%). 93% of the patients received expert medical help within two hours after being bitten. Virtually all patients received the antivenom (99%), tetanus prophylaxis (98%) and antibiotics (97%). In addition, a large number of patients received antihistamines (73%) and corticosteroids (78%). Remarkably, 94% of the patients received the first dose of the antivenom within four hours after the bite. 77% of the patients received one dose of the antivenom, and 22% received two or three doses. A total of 783 doses of the antivenom were used on 540 patients, and only one case of anaphylactic shock was recorded (0.2%). Furthermore, three cases of serum sickness occurred (0.6%). Of the antibiotics used, penicillin-based products were most frequently administered. Virtually all patients received the tetanus prophylaxis, and no tetanus cases occurred. 17% of the cases required surgical intervention, which most frequently included the incision of hemorrhagic blister (12%), then early debridment of superficial necrosis (3.2%) and fasciotomy (1.8%) (Figure 5). All of our surgically treated patients recovered successfully.

Discussion

In southern Croatia snakes are active from the end of winter to late autumn[5]. A majority of the bites were recorded in May and in summer months (June, July, August) because these are the months when the vipers are most active, and when people are very much engaged in agricultural activities in southern Croatia. Other European countries show a similar seasonal trend in the frequency of bites, but there is a difference with respect to the most hazardous month[1,3,11–13]. For example, in southern Croatia the highest number of bites happened in May, whereas in Spain this was in June, and in Italy in August[12,13]. *V. ammodytes* bites in this region were much more common than *V. berus* bites, because *V. ammodytes* is very widespread, whereas *V. berus* has only a very limited distribution confined to the peripheral regions of the county[5]. *V. ammodytes* is especially dangerous when climbing trees in search of food because of a high risk of inflicting bites on the head and the neck[5,6]. The bites on the neck and the head, as well as the bites directly into a blood vessel, are extremely dangerous. The injection of the venom into a blood vessel commonly leads to toxic shock, which may easily prove fatal, although this is not the rule[5,7]. The bites on the upper extremity took place more often than the bites on the lower extremity, mostly because of traditional agricultural work procedures, which involve the use of bare hands and traditional, non-mechanized equipment. Swiss, Italian and Spanish studies also found the bites on the upper extremity to be more frequent than the bites on the lower extremity. On the contrary, a Swedish study found the bites on the lower extremity to be more common[3,11,12,14]. The bites occurred most frequently in children, adolescents and older individuals, which was also found to be true in other studies[5,11,15]. Children and adolescents were frequently bitten while playing outdoors (in fields), and older individuals were bitten while being engaged in agricultural activities.

The clinical symptoms that appear following the bite of *V. ammodytes* or *V. berus* are mainly a result of the hematotoxic effects of the venom, and only to a smaller degree of the neutrotoxic effects[5,13]. All of the patients had swelling and over 90% had discoloration of skin and ecchymosis. Swelling, skin discoloration and ecchymosis, which occur soon after the venom is injected (within 4 hours), are used to distinguish a venomous from a non-venomous bite[5,16].

Massive swelling at the bite site leads to increased tissue pressure within the closed bonefascial space on the extremity, which may result in CS[5,7,17,18]. This is usually the case in children because of a larger quantity of venom per unit of body weight. In determining clinical diagnosis of CS and deciding on the use of fasciotomy we should be utterly cautious, because the surgical intervention itself can cause complications and permanent disability. Fasciotomy should only be carried out in patients with clinical symptoms and signs of compartment syndrome (painful passive stretch, hypoesthesia, compartment tenseness, and weakness), and measured compartment pressure over 30 mmHg (provided the instrument for measuring pressure is available). Fasciotomy should never be performed prophylactically[7,15,18,19]. In other studies on Euro-

pean viper bites, CS was either not recorded at all or it was found very rarely[1,3,8,9,12,19].

In this study in which *V. ammodytes* bites were predominant, there was a fairly large number of patients with clinically manifested neurological disturbances as a result of the neurotoxic effects of the venom. Specifically, 87 (16%) of our patients had cranial nerve paresis or paralysis (ptosis, ophthalmoplegia, dysphagia), and 6 (1.1%) of our patients had CNS depression only, without shock, hypotension, or cranial nerve paresis or paralysis. This indicates that the venom not only has direct neurotoxic action on cranial nerves, but it is also likely to have a primary toxic action on CNS as well.

The average duration of hospitalization in our study was longer than the averages reported in other studies[11,12], and it was correlated with the severity of envenomation. The frequency of moderate and severe envenomation in our study was much higher than that reported in other studies. For example, we report 44% of moderate and severe cases of envenomation, whereas the Swedish authors reported 27% of such cases, Italian 23% and French 25%[8,11,12]. A higher frequency of moderate and severe cases of envenomation in this study could be attributed to the fact that most bites in southern Croatia were inflicted by *V. ammodytes*, which delivers more venom and causes more severe envenomation than *V. berus* and *V. aspis* do.

The envenomation by *V. ammodytes* and *V. berus* is a medical emergency that requires urgent treatment, including first aid in the field and expert treatment at a medical center. In the field, the victim should be reassured, the bite site cleaned and the limb immobilized. Most importantly, the victim must be transported to the nearest medical center as soon as possible[5,6,16]. Older practices such as applying a tourniquet, making incisions at the bite site and sucking the venom out are no longer recommended[16]. At a medical center, the extent of swelling should be closely observed and monitored as well as the development of other symptoms and signs of envenomation. If the swelling spreads rapidly, or if the patient develops systemic signs of envenomation (shock, unconsciousness, severe gastrointestinal symptoms), antivenom should be administred[5,7,16]. In our region we recommend giving antivenom to patients with moderate and severe envenomation at the medical center. It should not be given to patients having mild and minor envenomation[13,14,16,20–22]. The antivenom should be administered intravenously[13,16,20]. The efficacy of the antivenom is the highest if it is administered within 4 hours after the bite, and in which case one dose of the antivenom is usually sufficient (10 ml). If envenomation is severe, two to four antivenom doses need to be used (20–40 ml)[20]. We used only Zagreb antivenom, and found complications following its administration to be very rare. In contrast, some authors found that complications were not uncommon following the use of Zagreb antivenom and other equine antivenoms[3,9,12,13,23].

Following the treatment with antivenom, tetanus prophylaxis should be administered, depending on the immunological status of the patient to tetanus[4,13,24]. In the past, a number of authors recommended the use of antibiotics prophylactically after the snakebite in all cases[6,13,25], but this is no longer recommended[4]. Antibiotics should only be given in the event of infection (according to the identity of the pathogen and its susceptibility to the antibiotic)[4]. The use of antihistamines and corticosteroids, previously advocated by some authors, is no longer recommended[1,5,13,27,28]. Corticosteroids are only used to treat allergic reactions to the serum, and some authors use them to treat CS[1,6,19,28.] If the patient is in shock or his or her general condition is severely altered, he or she should be transported to the intensive care unit, where all the necessary treatments need to be carried out, including the administration of infusion solutions, blood, sedatives, anticonvulsants, mechanical ventilation, hemodialysis, surgical intervention, etc[7–9,13,16,17,29].

In this study, the antivenom and particularly antibiotics, antihistamines and corticosteroids were used more often than it was suggested by the symptoms present. However, the use of these medicines should be limited to cases in which sufficient indication for their use exists, as it is described in the text.

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UGRIZI ZMIJA OTROVNICA U JUŽNOJ HRVATSKOJ

S A Ž E T A K

U retrospektivnoj studiji prikazana su 542 bolesnika otrovana zmijskim otrovom u južnoj Hrvatskoj, koji su liječeni u Kliničkoj Bolnici Split kroz 21-godišnje razdoblje. Cilj rada bio je utvrditi incidenciju ugriza otrovnica u južnoj Hrvatskoj, te prikazati epidemiološka obilježja, kliničku prezentaciju i načine liječenja otrovanja zmijskim otrovom na ovom području. Prosječna incidencija ugriza otrovnica u južnoj Hrvatskoj iznosila je 5,2/100 000 stanovnika godišnje. Ugrizi poskoka (*Vipera ammodytes*) su bili češći od ugriza riđovke (*Vipera berus*). Od ugriza su stradavale sve dobne skupine (od 1–82 godina), a najčešće (46%) stariji od 50 godina, te djeca i adolescenti u dobi od 0–19 godina (27%). Većina ugriza dogodila se tijekom toplih mjeseci proljeća i ljeta, najčešće u svibnju (22%). Od ugriza otrovnica uglavnom je stradavalo seosko pučanstvo obavljajući poljoprivredne radove. Ugrizi u ruku su bili češći (57%) nego ugrizi u nogu (42%). Prema težini otrovanja bilo je 15,1% bolesnika s vrlo blagim oblikom otrovanja, 40.5% s lakim, 26 % sa srednje teškim i 18% s teškim oblikom otrovanja. Smrtno su stradala dva bolesnika (0,4%). Gotovo svi bolesnici su primili antiviperini serum (proizvođač Imunološki zavod u Zagrebu), a komplikacije nakon njegove primjene su bile rijetke. Antiviperini serum se koristio češće nego je za to postojala stvarna indikacija.