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Toxicokinetics and correlation of carbamazepine salivary and serum concentrations in acute poisonings

Toksikokinetika i korelacija koncentracija karbamazepina u salivi i serumu kod akutnog trovanja

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Abstract

Background/Aim. Saliva is a body fluid which, like serum, can be used for determination of concentrations of certain drugs, both in pharmacotherapy as well as in acute poisonings. The aim of this study was to determine carbamazepine concentrations in both saliva and serum in acute poisoning in order to show if there is a correlation between the obtained values, as well as to monitor toxicokinetics of carbamazepine in body fluides. **Methods.** Saliva and serum samples were obtained from 26 patients treated with carbamazepine and 20 patients acutely poisoned by the drug immediately after their admission in the Emergency Toxicology Unit. Determination of salivary and serum carbamazepine concentrations was performed by the validated high pressure liquid chromatography-ultraviolet (HPLC-UV) method. **Results.** A significant correlation of salivary and serum carbamazepine concentrations in both therapeutic application and acute poisoning (r = 0.9481 and 0.9117, respectively) was confirmed. In acute poisonings the mean ratio between salivary and serum concentrations of carbamazepine (0.43) was similar to the mean ratio after its administration in therapeutic doses (0.39), but there were high inter-individual variations in carbamazepine concentrations in the acutely poisoned patients, as a consequence of different ingested doses of the drug. In acute poisoning the halftime of carbamazepine in saliva and serum was 12.57 h and 6.76 h, respectively. **Conclusion.** Our results suggest a possible use of saliva as an alternative biological material for determination of carbamazepine concentrations in therapeutic application and acute poisoning as well, and a possible extrapolation of the results obtained in saliva to serum concentrations of carbamazepine.

Key words:
carbamazepine; pharmacokinetics; poisoning; serum; saliva; chromatography; sensitivity and specificity.

Apstrakt

Uvod/Cilj. Slično serumu, saliva je biološki materijal koji se može primeniti za određivanje koncentracije lekova kako nakon terapijske primene, tako i u akutnom trovanju. Cilj ovog rada bio je da se odrede koncentracije karbamazepina u salivi i serumu u akutnom trovanju da bi se pokazalo da li postoji korelacija između dobijenih vrednosti, kao i da se isprati toksikokinetika karbamazepina u salivi i serumu. **Metode.** Uzorci salive i seruma uzeti su od 26 bolesnika na terapiji karbamazepinom i 20 bolesnika akutno otrovanih ovim lekom nakon prijema u toksikološku ambulantu. Određivanje koncentracije karbamazepina vršeno je validovanom metodom visokoefikasne tečne hromatografije sa ultravioletnom detekcijom (HPLC-UV). **Rezultati.** Potvrđena je značajna korelacija koncentracija karbamazepina u salivi i serumu nakon terapijske primene (r = 0,9481), kao i u akutnom trovanju ovim lekom (r = 0,9117). Prosečni odnos koncentracija karbamazepina u salivi i serumu u akutnim trovanjima (0,43) bio je sličan odgovarajućem parametru nakon terapijske primene leka (0,39), ali je bilo većih interindividualnih razlika u koncentracijama leka u akutnim trovanjima, zbog, najverovatnije, razlika u ingestiranim dozama karbamazepina. U akutnim trovanjima poluvreme eliminacije karbamazepina u serumu bilo je 12,57 h, a u salivi 6,76 h. **Zaključak.** Dobijeni rezultati govore o mogućoj primeni salive kao biološkog materijala za određivanje koncentracije karbamazepina tokom terapijske primene i u akutnom trovanju, kao i o mogućoj ekstrapolaciji vrednosti koncentracija karbamazepina u salivi na serumske koncentracije ovog leka.

Ključne reči:
karbamazepin; farmakokinetika; trovanje; serum; pljuvačka; hromatografija; osetljivost i specifičnost.

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Introduction

Traditional biological materials for qualitative and quantitative measurements of most drugs are serum and urine. Many substances and their metabolites are present in different concentrations in these samples. A number of data shows that saliva is a suitable alternative for determining plasma levels of many drugs. It has the advantage of noninvasive and easy sampling, so detection of drugs in saliva can be very useful [1–5].

Under standardized and well-controlled sampling conditions, therapeutic drug monitoring of anticonvulsant drugs in saliva can be useful for determining compliance with medication, especially in pediatric patients [6].

Correlation of different drugs concentrations given in therapeutic doses in saliva and blood are known. But, there is relatively insufficient data about drugs determination, and correlation of their concentrations in blood and saliva in cases of acute poisonings by them.

Since carbamazepine is the most used drug for treating seizures, it is understandable why it is also the most frequently used antiepileptic in self poisoning [7].

The aim of this investigation was to show if there is a correlation between carbamazepine salivary and serum concentrations and dynamic of their changes in the time.

Methods

Saliva and serum samples were collected from 20 patients acutely poisoned by carbamazepine after their admission in the Emergency Toxicological Unit. Every acutely poisoned patient had previously used carbamazepine for therapy.

Twenty six saliva and serum samples were also collected from the epileptic patients treated by carbamazepine at least 6 months. Sampling of saliva and blood were done 4 h after taking the drug.

Samples were stored at -20°C until preparation and analyzing. They were prepared and analyzed by the validated high pressure liquid chromatography-ultraviolet (HPLC-UV) method previously described by Djordjevic et al. [8].

Calculation of a saliva/serum ratio and linear regression dependence of carbamazepine concentrations in saliva and serum were done by the computer software Microsoft Excel 2003.

A correlation of carbamazepine concentrations in saliva and serum was determined by Pearson's regression analysis, and calculating its halftime in saliva and serum in acute poisonings was determined using the computer software WinNonlin.

Results

In the patients on a long-term therapy with carbamazepine, salivary and serum concentrations are shown in Table 1.

Salivary and serum carbamazepine concentrations in the acutely poisoned patients and their ratios are shown in Table 2. Carbamazepine salivary and serum concentrations during the followed time period are shown in Table 3. Carbamazepine saliva levels correlated well with blood concentrations. The salivary and serum concentrations ratio for different patients also correlated well.

In both cases of treated and acutely poisoned patients, carbamazepine concentrations in saliva had a lower value compared with those in serum when the samples were collected at the same time. The mean concentrations of carbamazepine in serum and saliva samples of treated patients were 4.40 mg/L (from 1.51 to 7.52 mg/L) and 1.71 mg/L (from 0.58 to 2.82 mg/L), respectively.

Table 1

Carbamazepine serum and saliva concentrations (c) and their ratios during therapeutic use of the drug

Patient N°	Serum (c) (mg/L)	Saliva (c) (mg/L)	Saliva/serum ratio	Patient N°	Serum (c) (mg/L)	Saliva (c) (mg/L)	Saliva/serum ratio
1	4.71	1.65	0.351	14	7.52	2.54	0.338
2	1.80	0.70	0.389	15	3.87	1.69	0.438
3	2.10	1.10	0.524	16	5.84	1.98	0.338
4	5.20	1.99	0.383	17	2.99	0.86	0.288
5	4.98	1.90	0.382	18	5.45	2.37	0.435
6	2.89	1.40	0.484	19	2.85	1.00	0.353
7	6.89	2.82	0.409	20	6.01	2.33	0.387
8	5.65	2.10	0.371	21	1.51	0.61	0.406
9	4.58	1.69	0.369	22	2.47	1.13	0.460
10	6.92	2.74	0.396	23	2.42	0.58	0.240
11	4.50	2.12	0.471	24	2.78	1.16	0.417
12	4.44	1.58	0.356	25	3.57	1.30	0.363
13	5.74	2.54	0.442	26	3.83	1.50	0.391
	Serum (c) (mg/L),		$\bar{x} \pm SD$: 4.29 ± 1.68				
	Saliva (c) (mg/L),		$\bar{x} \pm SD$: 1.67 ± 0.66				
	Saliva/serum ratio,		$\bar{x} \pm SD$: 0.391 ± 0.060				

For monitoring carbamazepine toxicokinetics, serum and saliva samples were taken in the subgroup of 6 patients immediately after their admission in the Emergency Toxicological Unit, as well as after 2.5 h and 4.5 h. Saliva samples were taken with a buffer impregnated by 3% citric acid.

In the acutely poisoned patients the mean serum and saliva concentrations were 9.54 mg/L (ranged from 1.24

Table 2
Carbamazepine serum and saliva concentrations (c) and their ratios in acute poisonings by the drug

Patient N°	Serum (c) (mg/L)	Saliva (c) (mg/L)	Saliva/serum ratio
1	4.61	0.31	0.067
2	2.41	0.87	0.361
3	6.02	3.84	0.638
4	25.03	7.14	0.285
5	30.30	7.26	0.239
6	1.24	0.16	0.129
7	5.55	3.47	0.625
8	5.02	2.37	0.472
9	12.86	4.72	0.367
10	7.34	2.46	0.335
11	16.78	5.81	0.346
12	3.28	2.35	0.716
13	9.76	3.66	0.375
14	3.34	2.50	0.748
15	23.70	12.48	0.527
16	29.30	14.71	0.502
17	12.71	7.10	0.559
18	35.77	19.49	0.545
19	28.35	12.56	0.443
20	10.36	4.52	0.436
Serum (c) (mg/L),	$\bar{x}$ ± SD: 14.28 ± 10.99		
Saliva (c) (mg/L),	$\bar{x}$ ± SD: 5.89 ± 5.19		
Saliva/serum ratio,	$\bar{x}$ ± SD: 0.436 ± 0.180		

Table 3
Changes in carbamazepine serum and saliva concentrations (c) in patients acutely poisonet by the drug within 6 h after hospital admission

Patient N°	Time after admission (h)	Serum (c) (mg/L)	Saliva (c) (mg/L)	Saliva/serum ratio
	0	23.70	12.48	0.526
1	2.5	19.45	7.96	0.410
	4.5	16.07	6.39	0.397
	0	29.30	14.71	0.502
2	2.5	27.30	11.33	0.415
	4.5	21.91	9.82	0.448
	0	12.71	7.81	0.614
3	2.5	11.12	6.06	0.545
	4.5	9.89	5.70	0.576
	0	35.77	19.49	0.545
4	2.5	27.60	11.97	0.434
	4.5	26.22	9.96	0.380
	0	28.35	12.56	0.443
5	2.5	25.68	10.61	0.413
	4.5	21.83	8.40	0.385
	0	10.36	4.52	0.436
6	2.5	9.43	3.43	0.364
	4.5	8.01	2.62	0.327
Serum (c) (mg/L),		$\bar{x}$ ± SD: 20.26 ± 8.42		
Saliva (c) (mg/L),		$\bar{x}$ ± SD: 9.21 ± 4.24		
Saliva/serum ratio,		$\bar{x}$ ± SD: 0.453 ± 0.080		

mg/L to 30.30 mg/L) and 3.35 mg/L (ranged from 0.16 mg/L to 7.26 mg/L), respectively.

The carbamazepine saliva/serum ratio after therapeutic application was 0.39, and in acute poisonings 0.43. In both cases, *ie.* in therapeutic use and acute poisonings, there was a strong correlation between carbamazepine salivary and serum concentrations. The coefficients of correlation were 0.9481 and 0.9117 ($p < 0.005$), respectively.

The serum and saliva halftimes of carbamazepine in acute poisonings were 12.57 h and 6.7 h, respectively. On the other hand, when the drug was used in therapeutic doses, its serum and saliva halftime was in a wide range from 10 to 35 h, and 4.1 to 33.1 h, respectively.

The correlation between carbamazepine serum and saliva concentrations was calculated by using Pearson's regression analysis ($y = 1.03*x - 0.897$; $r = 0.9427$, $p < 0.01$).

Discussion

It is known that saliva can be suitable medium for monitoring free concentrations of carbamazepine providing a noninvasive method of sampling biological material for drug determination. Monitoring of carbamazepine in saliva is of particular interest in the management of children with epilepsy, and in geriatric patients in whom trombosed peripheral veins might limit blood sampling [9].

For determination of salivary carbamazepine concentrations when the drug is given in therapeutic doses stimulated or non-stimulated saliva can be used [10]. Age, gender and time of sampling have no influence on carbamazepine proteins binding and its saliva serum ratio [11].

Findings of Rosenthal et al. [10] have shown that stimulation of salivary excretion has no influence on carbamazepine concentration.

Salivary carbamazepine concentrations were independent of volume of fluid produced, pH of saliva, and degree of stimulation [12].

Gorodisher et al. [13] used stimulated saliva for monitoring carbamazepine concentration in children. Their results showed that correlations between saliva and free plasma anticonvulsant concentrations were equal or only slightly better than between saliva and plasma total concentrations.

Significant linear relationships between saliva and total plasma concentrations and between saliva and free plasma concentrations were observed for carbamazepine. However, salivary concentrations of carbamazepine were significantly more reliable as predictors of their respective free plasma concentrations than of their respective total plasma concentrations. It is considered that measurement of carbamazepine in saliva of chronically medicated epileptic patients provides a more reliable estimate of pharmacodynamically active, free concentrations of this compound in plasma [11].

We also used stimulated saliva for carbamazepine monitoring. For salivary stimulation we used, like many other authors, citric acid. There is a strong correlations between salivary and serum concentrations of carbamazepine ($r = 0.9481$) in accordance with literature data. For example in the study of Vasudev et al. [5] the coefficient of correlation was $r = 0.659$, in that of Rozenthal et al. [10] $r = 0.89$, al Za`abi et al. [14] $r = 0.99$ and Knot and Reynolds [15] $r = 0.94$.

Ratios between carbamazepine saliva and total blood concentrations presented by different authors, were in the range from 0.27 to 0.386 [10, 15, 16]. The described ratios show a level of carbamazepine protein binding, because the salivary drug concentration is equal to serum free concentration. This is proved by the results presented by al Za`abi et al. [14], where the mean ratio of carbamazepine saliva/free serum concentration was 1.02 +/- 0.11.

Our results are similar to those of other authors. The carbamazepine saliva/serum ratio was 0.39, which means that concentration of carbamazepine in saliva was about 39% of the total blood concentration.

We also studied a saliva/serum ratio in acute poisonings by carbamazepine. Data about correlation of salivary and serum concentration in poisoning by antiepileptic drugs do not exist.

As in patients on therapy with carbamazepine, in those overdosed with the drug, we also used stimulated saliva. Stimulating of salivation enables sampling in patients who are in coma, dehydrated or have insufficient vein pathway.

We found that as in carbamazepine treatment, there was a strong correlation between saliva and serum concentrations of carbamazepine in cases of drug poisoning ($r = 0.9117$, $p < 0.05$). The salivary and serum ratio of carbamazepine concentrations was slightly higher (0.43) than in a long-term use of therapeutic doses carbamazepine (0.39). A higher saliva/serum ratio in acute poisoning could be explained by increasing of free serum carbamazepine concentration due to saturation of finding proteins after ingestion of high doses of drug.

There were high inter-individual variations, as a result of poisoning with different doses of carbamazepine, various times from ingestion to admission to the Emergency Toxicological Unit and various time from ingestion to sampling.

The results concerning dynamics of carbamazepine concentrations changes in time showed decreasing of the carbamazepine saliva/serum ratio during the observed period. It could be explained by metabolism of free serum carbamazepine, reflected by decreasing saliva concentration.

We also calculated a value of carbamazepine halftime in acute poisonings. Halftime is a parameter which enables predicting a degree of drug elimination and ingoing to therapeutical range. Our data indicate that in acute poisoning the halftime of carbamazepine in serum and saliva is 12.57 h, and 6.76 h, respectively.

The halftime of carbamazepine in serum after its long-term treatment is in range from 10 h to 35 h [17]. Data about carbamazepine saliva halftime were in wide range from 4.1 h to even 33.1 h [18].

Our results were similar to those in the literature because all of the poisoned patients had carbamazepine in their therapy for long time.

Because there is a strong correlation between salivary and serum concentrations in acute poisonings, the salivary concentrations could be used for calculating carbamazepine serum concentrations by using the formula $y = 1.03*x - 0.897$, where y is a serum concentration of carbamazepine, and x is a salivary one.

Conclusion

The results of this study demonstrate that monitoring of salivary carbamazepine concentrations can be a realistic alternative to blood in routine clinical analysis after therapeutic application or in acute poisoning. Saliva is an attractive alternative biological material, due to its painless collection and noninvasive sampling comparing with blood. Salivary drug levels correlated well with serum drug concentrations in acute poisoning. Moreover, it is possible to extrapolate concentrations of carbamazepine obtained from saliva to their serum concentrations on basis of given correlation curves in patients acutely poisoned with this drug.

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