

ISOTHERMAL AND NON-ISOTHERMAL KINETICS OF HYDROLYSIS OF 1-[2-(2-PENTYLOXYPHENYLCARBAMOYLOXY)-(2-METHOXYMETHYL)-ETHYL]-PERHYDROAZEPINIUM CHLORIDE (BK 129)

IZOTERMICKÁ A NEIZOTEREMICKÁ KINETIKA HYDROLÝZY 1-[2-(2-PENTYLOXYFENYLKARBAMOYLOXY)-(2-METOXYMETYL)-ETYL]-PERHYDROAZEPÍNÍUMCHLORIDU (BK 129)

Original research article

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Abstract The substance BK 129 - 1-[2-(2-pentyloxyphenylcarbamoyloxy)-(2-methoxymethyl)-ethyl]-perhydroazepinium chloride was prepared in terms of influence of the connecting chain between the carbamate functional group and the basic part of molecule on biological activity. Such a structural feature is important with regard to its stability. In this work we determined the rate constants of alkaline hydrolysis of this compound at increased temperature under isothermal and non-isothermal conditions. The hydrolysis was also performed in buffer solutions with the purpose of evaluating its stability. Non-isothermal tests of stability enable to reduce the number of analyses. The necessary data for stability of compound are in this way achieved in a short time.

Slovak abstract Látka BK 129 - 1-[2-(2-pentyloxyfenylkarbamoyloxy)-(2-metoxymetyl)-etyl]-perhydroazepíniumchlorid bola pripravená v rámci štúdia vplyvu spojovacieho reťazca medzi karbamátovou funkčnou skupinou a základnou časťou molekuly na biologickú aktivitu. Táto štruktúrna zložka je dôležitá ohľadom na stabilitu. V tejto práci sú stanovené rýchlostné konštanty alkalického hydrolyzy tejto látky za zvýšenej teploty za izotermických a neizotermických podmienok. Hydrolyza látky bola tiež vykonaná v tlmivých roztokoch kvôli vyhodnoteniu jej stability. Neizotermické testy stability umožňujú redukovat počet analýz. Potrebné údaje o stabilite látky sa týmto spôsobom dajú dosiahnuť v krátkom čase.

Keywords antiarrhythmics – stability study – kinetics of hydrolysis – non-isothermal kinetics

Kľúčové

slová: antiarytmiká - štúdia stability - kinetika hydrolyzy - neizotermická kinetika

INTRODUCTION

The novel potential drug substance BK 129 (Fig. 1) (Búčiová et al., 1987) possesses the biological activity up to five times higher than that of lidocaine by the antagonisation of ventricle extrasystoly, fibrillation and mortality, and underwent a detailed pharmacological study (Gibala et al., 1987, 1987). For these reasons, the analytical profile of the substance BK 129 was also elaborated (Sedlárová et al., 1995). The aim of this work was to determine the rate constants of alkaline hydrolysis of substance BK 129 at increased temperature in sodium hydroxide solution $c = 0.1 \text{ mol/l}$, as well as the study of its hydrolysis in buffer solutions with the purpose of evaluating its stability. In alkaline media, the basic esters of phenylcarbamic acid undergo the hydrolysis and decompose to substituted aniline, basic

alcohol and carbon dioxide. The studied substance possesses the branched connecting chain which differentiates it from the other derivatives, e.g. heptacaine (Čižmárík et al., 1976).

In the preliminary study of alkaline hydrolysis of compound BK 129 at several temperatures, we have observed that the substance was decomposed very quickly up to a certain degree, but later the rate of decomposition reaction slowed down and the reaction rate reached an equilibrium character. At lower temperatures, the alkaline hydrolysis was very slow with identical course. Therefore, we proceeded to solve this problem by a non-isothermal kinetic study also, which is at present successfully used in the study of stability of drugs (Junnakar&Stavchansky, 1995, Lee et al., 1998, Ficara et al., 1999).

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In the case of non-isothermal processes, the temperature is not constant but changes with time. The first mathematic exact non-isothermal accelerated test of stability was introduced by Rogers (Rogers, 1965) by using the temperature/time function:

$$\frac{1}{T_0} - \frac{1}{T_t} = B \ln(1 + t) \quad (1)$$

where T_0 is the thermodynamic temperature in K at the start of experiment, T_t is the temperature in the time t , B is the constant of rate of proportionality which can be chosen as desired, and t is the time. If the equation (1) is applied to Arrhenius equation, it will be for the 1st order reaction obtained in the relationship:

$$\ln(\ln c_0/c_t) = \ln k_0 - \ln(1 + E_A B/R) + (1 + E_A B/R) \ln(1 + t) + \ln[1 - (k_0/k_t)^{1+R/E_A B}] \quad (2)$$

where c_0 is the concentration of substance in the time $t = 0$, c_t is the concentration in the time t , k_0 is the rate constant in the time $t = 0$, k_t is the rate constant in the time t , E_A is the activation energy, and R is the universal gas constant (8.315 J/mole/K). The last member of equation may be omitted when k_t rises. The graphic expression of function $\ln(\ln c_0/c_t) = f(\ln(1 + t))$ is the straight line. The slope of this function is equal to $(1 + E_A B/R)$. Because the constants B and R are known, it is possible to count the activation energy E_A in this way. The line will intersect the ordinate at $\ln k_0 - \ln(1 + E_A B/R)$. The decomposition rate constant k_0 at the temperature T_0 can be calculated. In the non-isothermal test by Eriksen and Stelmach during the experiment, the temperature is increased hyperbolically (Eriksen&Stelmach, 1965):

$$\frac{1}{T_0} - \frac{1}{T_t} = B \cdot t \quad (3)$$

If the equation (3) is applied into Arrhenius equation it will be for the 1st order reaction obtained in the relationship:

$$\ln(\ln c_0/c_t) = (E_A \cdot B \cdot t)/R + \ln[k_0 \cdot R/E_A \cdot B(e^{E_A \cdot B \cdot \Delta t/R} - 1)] \quad (4)$$

Plotting values of $\ln(\ln c_0/c_t)$ vs time t obtained a straight line where the slope equals $E_A \cdot B/R$ and the second summand equal the intercept of ordinate. The equation is dependent on the condition that the intervals between measurements are the same, because the value of Δt must be constant (Kersten&Göber, 1984).

MATERIALS AND METHODS

Studied compounds

The studied compound was synthesised as hydrochloride by authors [4] and is depicted in Fig. 1. All other chemicals and solvents were of analytical reagent grade.

Apparatus

The Thermostat Memmert WB 10 (Germany), spectrophotometer Spectronic 20 D, Milton Roy (Germany) and spectrophotometer 8452 A DIODE ARRAY Hewlett Packard (USA).

Kinetics of hydrolysis

The hydrolysis of compound BK 129 was carried out in aqueous ethanolic buffer solutions at pH 7.0 and 8.0 (Wells, 1988) as well as in NaOH 0.1 mol/l (pH 13.0) in closed flasks tempered in ultra thermostat. The buffer solutions at pH 7.0 and 8.0 possessed an amount of NaOH in the concentration of 0.029 mol/l and 0.046 mol/l, respectively. The concentration of KH_2PO_4 in both buffer solutions was 0.05 mol/l. The ionic strength of buffers was 0.1 mol/l; it was achieved by adding the appropriate amount of KCl. The $\text{C}_2\text{H}_5\text{OH}$ concentration was 50 % (V/V). The concentration of BK 129 was $6.993 \cdot 10^{-4}$ mol/l in buffers and $4.662 \cdot 10^{-4}$ mol/l in NaOH solution, respectively. The hydrolysis was carried out at $21.0 \text{ }^\circ\text{C} \pm 0.2 \text{ }^\circ\text{C}$, $37.0 \text{ }^\circ\text{C} \pm 0.2 \text{ }^\circ\text{C}$ in isothermal study and from $30 \text{ }^\circ\text{C}$ to $70.0 \text{ }^\circ\text{C}$ in non-isothermal study with

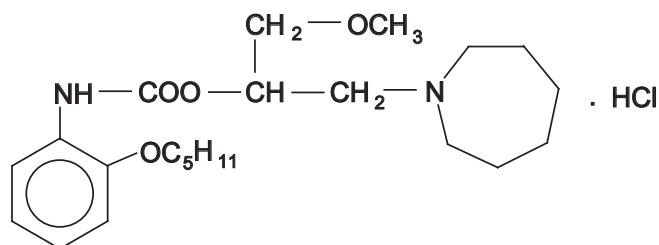


Fig. 1. Substance BK 129

rise of temperature 10 °C within 1 h. The hydrolysis at 70.0 °C ± 0.2 °C was carried out after attaining this temperature in non-isothermal study. The concentration of the 2-pentyloxylaniline, produced by the reaction, was determined spectrophotometrically in visible region using the diazotisation reaction with sodium nitrite and copulation with 2-naphthole (Stankovičová et al., 1975). The absorbance of solutions was measured at 492 nm.

Calculations

The rate constants of hydrolysis were calculated using the kinetic equation of the pseudo 1st order (Treindl, 1990):

$$\ln(b - x) = \ln b - k t \quad (5)$$

The half-life $t_{0.5}$:

$$t_{0.5} = \frac{0.693}{k} \quad (6)$$

The time during the stored solution contains 90 % of compound:

$$t_{0.9} = \frac{0.105}{k} \quad (7)$$

The values of activation energy E_A were determined by the Arrhenius equation:

$$k = Ae^{-E_A/RT} \quad (8)$$

where A is the preexponential member, R is the molar gas constant, T is the thermodynamic temperature, and E_A is the activation energy.

Values of E_A in the non-isothermal study were determined by the equations (2) and (4), respectively (Rogers, 1965, Eriksen&Stelmach,1965).

RESULTS AND DISCUSSION

In this paper the course of alkaline hydrolysis of substance BK 129 in aqueous ethanolic buffers solutions at equal ionic strength ($\mu = 0.1$ mol/l) as well as in aqueous ethanolic sodium hydroxide solution 0.1 mol/l was investigated. As reaction media for stability testing (Wells, 1988), the buffers were chosen with different concentrations of sodium hydroxide, where the presence of ethanol was required because of the low basic solubility of compound. In all cases, the ethanol concentration was equal to 50 % (v/v). The decomposition of substance BK 129 was firstly investigated in aqueous-ethanolic sodium hydroxide solution $c = 0.1$ mol/l at 40.0 °C and at 60.0 °C. The results of alkaline hydrolysis determined under isothermal conditions are given in Table 1. The substance decomposed very quickly up to a certain degree. The rate constants of pseudo 1st order are calculated from linear part of straight line because later the rate of decomposition reaction slowed down and the reaction rate reached an equilibrium.

Table 1. Results of kinetics of alkaline hydrolysis study of BK 129 at 40.0 and 60.0 °C in sodium hydroxide solution 0.1 mol/l

Temperature	k	$t_{0.5}$	n	r	F	s	a_0	a_1
°C	[h ⁻¹]	[h]						
40.0	$1.162 \times 10^{-2} \pm 5.91 \times 10^{-4}$	59.6	14	0.985	385.8	0.00442	0.00429	0.01162
60.0	$4.733 \times 10^{-2} \pm 3.38 \times 10^{-3}$	14.6	5	0.992	390.3	0.00651	-0.00161	0.04733

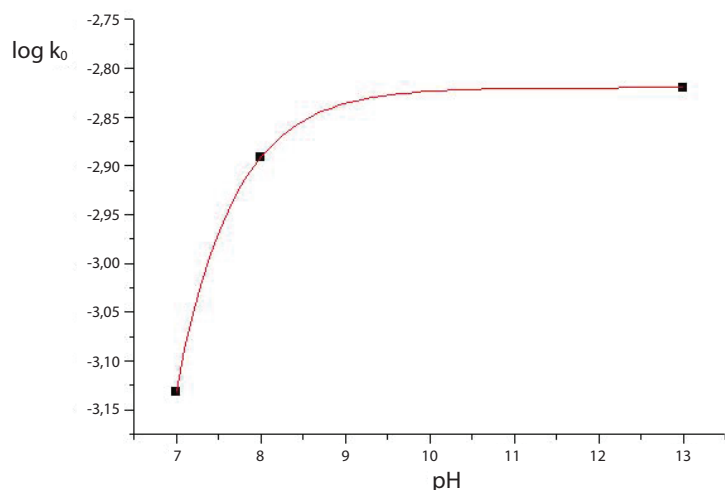


Fig. 2. Dependences of $\log k$ of compound BK 129 vs pH at 37 °C determined by isothermal study, the $\log k - \text{pH}$ profile

In order to decrease the reaction rate, the course of hydrolysis was observed at the laboratory temperature of 21.0 °C, and by accelerated isothermal stability test at 37.0 °C and at 70.0 °C in buffer solutions. In Table 2, the results of isothermal study are presented. The rate constants of pseudo 1st order rise with concentration of sodium hydroxide in buffers, as well as with temperature. For the study of stability, the values $t_{0.9}$ in days calculated for hydrolysis at 21.0 °C are important, which express the time during the stored solution that contains 90 % of compound. The results show that substance BK 129 is most stable in solution with pH 7, where the value $t_{0.9}$ equals 184 days. Figure 2 shows the dependences of $\log k$ of compound BK 129 on pH at 37 °C determined by isothermal study, the $\log k$ – pH profile. In Table 3 are given the values of activation energy E_A of hydrolysis of substance BK 129 in reaction solutions and parameters of straight lines. The reason of relatively high value of deviation of activation energy may be probably caused by too great a difference between temperatures used. The value of activation energy determined in

sodium hydroxide solution 89.2 kJ · mol⁻¹ is comparable with the values determined for 2-hexyloxy substituted 1-methyl-2-piperidinoethyl ester of phenylcarbamic acid (Stankovičová et al., 1999).

The hydrolysis of compound BK 129 by non-isothermal conditions was performed in range from 30.0 °C to 70.0 °C, with rate of heating 10 °C within an hour (Stankovičová et al., 2009). The constant B was calculated from equation (1) with rise of temperature 10 °C/h. The calculated value of B was: $B = 2.49 \times 10^{-4} \text{ K}^{-1}$ (Fig. 3). The values of E_A were calculated from equation (2), by non-isothermal method of study; in the Table 4 are given the parameters of straight lines. The programmed increase of temperature was carried out from 30 °C to 70 °C and the E_A values were calculated from the two parts of straight line, firstly from range of temperatures from 30.0 °C to 50.0 °C and then from range 50.0 °C to 70.0 °C, respectively, because for determination of activation energy it is suitable to use the close interval of temperature. The value of k_0 represents the calculated rate constant for starting temperature. In buffers as well as in sodium

Table 2. Results of kinetics of isothermal hydrolysis study of stability in buffer solutions of substance BK 129

Temperature	pH	k	$t_{0.5}$	$t_{0.9}$	n	r	F	s	a_0	a_1
°C		[h ⁻¹]	[h]	[d]						
21	7	$2.373 \times 10^{-5} \pm 9.36 \times 10^{-7}$	29203	184	6	0.997	642.2	0.000119	8.43×10^{-5}	2.37×10^{-5}
21	8	$5.356 \times 10^{-5} \pm 1.85 \times 10^{-6}$	12939	82	8	0.996	838.2	0.000407	1.63×10^{-4}	5.36×10^{-5}
21	13	$4.537 \times 10^{-4} \pm 3.24 \times 10^{-5}$	1527.5	10	7	0.988	196.6	0.00546	-3.41×10^{-3}	4.54×10^{-4}
37	7	$5.688 \times 10^{-4} \pm 1.48 \times 10^{-5}$	1216	8	12	0.996	1484	0.000399	3.67×10^{-4}	5.69×10^{-4}
37	8	$9.918 \times 10^{-4} \pm 4.83 \times 10^{-5}$	699	4.4	8	0.993	421.0	0.000341	4.96×10^{-4}	9.92×10^{-4}
37	13	$1.728 \times 10^{-3} \pm 9.79 \times 10^{-5}$	401	2.5	12	0.984	311.9	0.00251	2.65×10^{-3}	1.73×10^{-3}
70	7	$2.015 \times 10^{-3} \pm 1.80 \times 10^{-4}$	344	2.2	5	0.988	124.6	0.000285	-6.80×10^{-4}	2.02×10^{-3}
70	8	$5.272 \times 10^{-3} \pm 6.06 \times 10^{-4}$	131	0.8	4	0.987	75.71	0.000677	-1.58×10^{-2}	5.27×10^{-3}
70	13	$5.428 \times 10^{-2} \pm 5.74 \times 10^{-3}$	12.8	0.08	7	0.968	89.35	0.0186	7.76×10^{-2}	5.43×10^{-2}

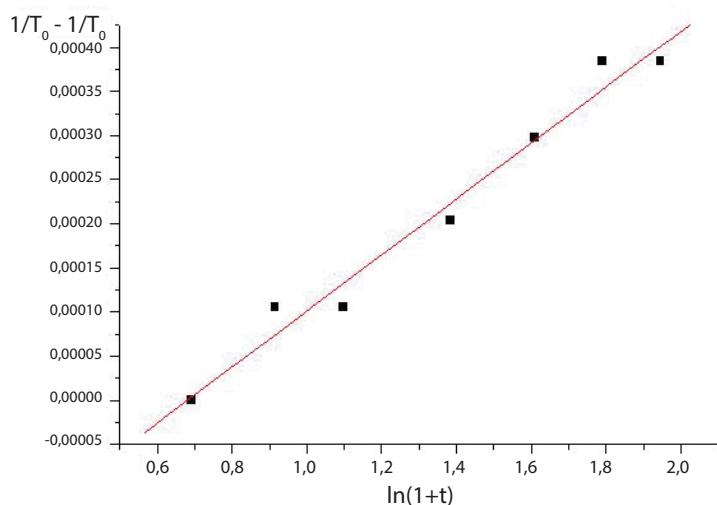


Fig 3. The rise of temperature

Table 3. Values of activation energy E_A of hydrolysis of substance BK 129 determined by isothermal study

pH	E_A [kJ·mol ⁻¹]	N	r	F	s	a_0	a_1
7.0	71.0 ± 30	3	0.921	5.56	0.5463	8.265	-3706.4
8.0	74.9 ± 24	3	0.951	9.39	0.4438	9.252	-3912.0
13.0	89.2 ± 9.6	5	0.983	86.57	0.1909	8.878	-4658.3

Table 4. Results of non isothermal study of hydrolysis of substance BK 129 determined by Rogers [10]

pH	Temperature interval °C	k_0 [h ⁻¹]	$t_{0.5}$ [h]	$t_{0.9}$ [day]	E_A [kJ·mol ⁻¹]	n	r	F	s
7.0	50 – 70	7.81×10^{-4}	887	5.6	31.9	5	0.999	14997	0.00611
8.0	30 – 50	1.284×10^{-3}	540	3.4	48.1	4	0.980	47.52	0.2362
13.0	30 – 70	1.552×10^{-3}	446	2.8	45.5	12	0.978	219.8	0.2204

hydroxide solutions, the values of E_A are lower than that determined by isothermal kinetics but the calculated rate constants for several temperatures are comparable with rate constants determined experimentally by isothermal kinetics of hydrolysis. The calculated rate constant of hydrolysis by non-isothermal method by Rogers is given in Table 5. In the Fig. 4 is depicted the course of hydrolysis of substance at pH 7 in the range of temperatures from 50.0 °C to 70.0 °C. The results of non-isothermal tests calculated by Eriksen and Stelmach method (Eriksen&Stelmach, 1965) are given in Table 6. The calculated value of B was: $B = 7.74 \times 10^{-5} \text{ K}^{-1}$. The values of activation energy determined in this way are comparable with that determined

by the non-isothermal study by Rogers, but the rate constant is not in consent with those determined by isothermal study as well as by other method given above. Non-isothermal tests of stability enable to reduce the number of analyses. The necessary data for stability of compound are in this way achieved in a short time but it is necessary to choose the suitable programme for temperature rise. It is also important to calculate the kinetic parameters only from the linear part of straight line of the studied relationship (Kersten&Göber, 1984).

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Table 5. Calculated rate constant of hydrolysis by non isothermal study of stability

pH	Temperature interval °C	k_{25} [h ⁻¹]	k_{37} [h ⁻¹]
7	50 – 70	3.37×10^{-4}	4.75×10^{-4}
8	30 – 50	9.33×10^{-4}	1.98×10^{-3}
13	30 – 70	1.15×10^{-3}	2.33×10^{-3}

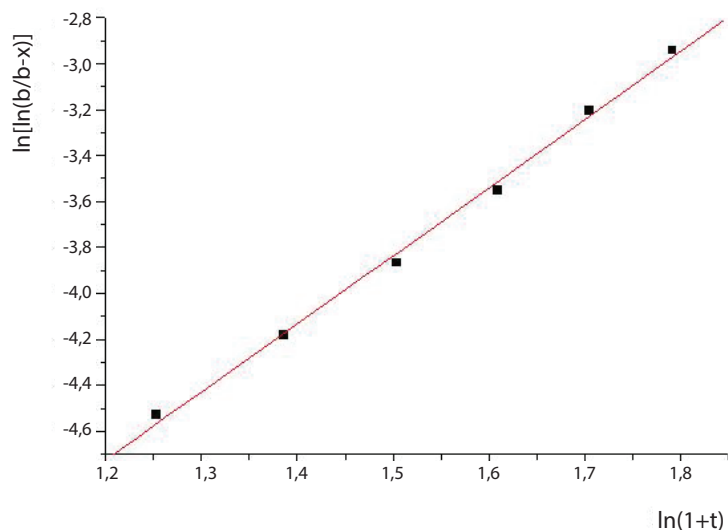


Fig. 4. Non isothermal test – the course of hydrolysis of substance BK 129 at pH 7

Table 6. Results of non isothermal study of hydrolysis of substance BK 129 determined by Eriksen and Stelmach [3]

pH	Temperature interval °C	a_0	a_1	E_A [kJ·mol ⁻¹]	n	r	F	s
7.0	30 – 70	-6.029	0.3334	35.8 × 1.4	9	0.994	287.0	0.0536
8.0	50 – 70	-5.619	0.3058	32.8 × 1.5	4	0.998	461.5	0.02362
13.0	30 – 70	-5.867	0.5802	51.2 × 1.9	9	0.995	694.7	0.08562

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