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Non-isothermal kinetics of benzo[*a*]pyrene photooxidation in glyceryl trioctanoate at various heating rates

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Summary

Benzo[*a*]pyrene (BaP) was dissolved in glyceryl trioctanoate, heated in a glass reaction vessel within a temperature range of 303.15–473.15 K at the heating rates 0.1, 0.2, 0.5, 1 and 2 $K \cdot min^{-1}$, respectively, and simultaneously exposed to UV light at the wavelength $\lambda = 365$ nm at the radiation power of 20 $mW \cdot cm^{-2}$ in order to study the decrease of BaP concentration due to its photooxidation. The reaction mixture was sampled at temperature increments of 10 K and the concentration of BaP was determined by HPLC with fluorescence detector. As observed, the BaP concentration decreased from initial 100 $\mu g \cdot l^{-1}$ in dependence on the heating rate. From the experimental results, the kinetic parameters of the first-order kinetic equation ($A = (2.8 \pm 5.9) \times 10^{-14}$ min^{-1}, $D = (5.86 \pm 0.43) \times 10^{-2}$ K^{-1}) describing the kinetics of BaP photooxidation were obtained. Knowing the kinetic parameters, modelling of the photooxidation for any time/temperature regime is feasible without a deeper insight into the mechanism of the process. The advantage of the multiple heating rate approach lies in the fact that a larger part of the kinetic hypersurface is mapped so that the kinetic parameters obtained are more reliable and trustworthy as when obtaining the kinetic parameters just from a single kinetic curve.

Keywords

polycyclic aromatic hydrocarbons; benzo[*a*]pyrene; non-isothermal kinetics; HPLC; photooxidation; glyceryl trioctanoate; heating

Polycyclic aromatic hydrocarbons (PAH) are permanent contaminants formed in various foods as a result of technological procedures carried out at elevated temperatures such as grilling, drying, frying and mainly smoking [1, 2]. For a long time, their content was believed to be constant in the food matrix, resistant to physico-chemical changes and environmental factors. However, it was found recently that their content may be affected by various technological procedures and interactions with packaging [3, 4]. Moreover, the content (and also toxicity) of these compounds may also be changed by solar radiation, which induces formation of reactive oxidized products [5, 6]. As known, PAH molecules contain two or more conjugated benzene rings that facilitate absorption of UV-A radiation (wavelength λ from 320 nm to 400 nm), UV-B radiation (λ from 290 nm to 320 nm) and also visible light (λ from 400 nm to 700 nm) in some instances. This may lead to photoactivation of PAH and to an increase in their toxicity via the photosensitized production of singlet oxygen and photomodification of original molecules that results in the formation of oxy-PAH compounds [7, 8]. Many products generated through photooxidation exhibit greater toxicity than the parent PAH and have a potential to generate toxic compounds that could negatively impact living systems and human health. On the other hand, photodegradation is an important transformation pathway for most PAH because this process preferentially attacks the same tertiary carbon atoms that tend to block biodegradation [7, 9]. As already proven, PAH deposited on a surface of smoked food are partially oxidized due to the presence of light and oxygen to form oxy-PAH compounds [10]. However, pho-

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tooxidation of PAH leads to formation of an enormous number of compounds hardly separable by current analytical techniques. In order to simplify the study of photooxidation processes, benzo[*a*]-pyrene (BaP) as a representative compound of PAH was photooxidized in a non-polar matrix in the presence of food antioxidants to characterise the physico-chemical process taking place during the photooxidation. As found, the reaction obeys the first order kinetics and the reaction itself is considerably accelerated by such important food antioxidants as BHT and guaiacol [11]. The aim of this work was to describe the non-isothermal kinetics of BaP photooxidation dissolved in glyceryl trioctanoate, exposed to UV radiation at $\lambda = 365$ nm, at various rates of heating.

MATERIALS AND METHODS

Chemicals

Benzo[*a*]pyrene (BaP) was of analytical grade, purchased from Supelco (Bellefonte, Pennsylvania, USA) in a solid state. Glyceryl trioctanoate was purchased from Sigma-Aldrich (Steinheim, Germany). Methanol of analytical grade was purchased from Merck (Darmstadt, Germany).

Experiments

BaP was dissolved in glyceryl trioctanoate to obtain the concentration of 100 μg·ml^{-1}. The solution was then placed in a metal block thermostat (Liebisch, Bielefeld, Germany) and heated from 303.15 K to 473.15 K at heating rates of 0.1, 0.2, 0.5, 1 and 2 K·min^{-1}. The temperature inside the 40 ml reaction vessel was monitored by a thermometer (Ecoscan Temp JKT temperature meter with Probe 3T520C; Eutech Instruments Europe, Nijkerk, the Netherlands). For HPLC analysis, a 100 μl of sample was taken at temperature increments of 10 K, diluted and injected into HPLC system. The experiments were carried out in duplicate.

HPLC conditions

HPLC analyses were carried out on the Shimadzu instrument (Kyoto, Japan). It consisted of solvent delivery module LC-20AD, autosampler SIL-20A, degasser DGU-20A5, column oven CTO-20A, communications bus module CBM-20A, diode array detector SPD-M20A and fluorescence detector RF-10AXL. The analytical separation was performed on Zorbax Eclipse XDB-C18 (50 mm × 4.6 mm, 1.8 μm) column (Agilent Technologies, Palo Alto, California, USA) using isocratic elution with methanol at a flow rate of 0.5 ml·min^{-1} at 35 °C. Fluorescence detector operated at an excitation wavelength of 300 nm and an emission wavelength of 410 nm.

Treatment of experimental results

Mechanisms of the processes in condensed phase are very often unknown or too complicated to be characterized by a simple kinetic model. They tend to occur in multiple steps that have various rates. In order to describe their kinetics, methods based on the single-step approximation are frequently used [12, 13]. The single-step approximation employs the assumption that the rate of a complex process can be expressed as a product of two separable functions independent of each other, the first one, $k(T)$, depending solely on the temperature T and the other one, $f(\alpha)$, depending solely on the conversion of the process, α. The rate of the complex multi-step condensed-state process thus can be formally described as [12–14]

$$\frac{\mathrm{d}\alpha}{\mathrm{d}t} = k(T)f(\alpha) \tag{1}$$

Eq. (2) is mostly called the general rate equation. Indeed, it resembles a single-step kinetics equation, even though it is a representation of the kinetics of a complex condensed-phase process. In general, kinetics of a complex process should be described by a set of rate equations. The single-step approximation thus resides in substituting the set of kinetic equations by the sole single-step kinetics equation [12–14].

The temperature function in eq. (1) is prevailingly expressed by the Arrhenius equation. In papers [12–15] it was justified that, due to complexity of the processes, the temperature function can hardly be considered the rate constant so that there is no reason to be confined to the Arrhenius temperature function. In papers [15, 16] it was demonstrated that the best agreement with experience, even in the case of extrapolation to lower temperatures, provides the following temperature function:

$$k(T) = Ae^{DT} \tag{2}$$

where A and D are parameters. If it is assumed that the conversion function can be characterized by n-th order kinetics, the rate equation is

$$\frac{\mathrm{d}\alpha}{\mathrm{d}t} = Ae^{DT}(1-\alpha)^n \tag{3}$$

Preliminary calculations showed that the best agreement between the experimental and calculated values of conversion was obtained for the reaction order $n = 1$. For this case, eq.(3) takes the form

$$\frac{\mathrm{d}\alpha}{\mathrm{d}t} = A e^{DT}(1-\alpha) \tag{4}$$

During the experiment, temperature obeys the linear relationship

$$T = T_0 + \beta t \tag{5}$$

where T_0 is the initial temperature, β is the heating rate and t stands for time. Combining eq. (4) and eq. (5), one can get:

$$\frac{\mathrm{d}\alpha}{\mathrm{d}t} = \frac{A}{\beta} e^{DT}(1-\alpha) \tag{6}$$

After separation of variables and integration, it can be obtained:

$$-\beta \ln(1-\alpha) = \frac{A}{D}\left(e^{DT}-1\right) \tag{7}$$

The lower integration limit in eq. (7) was set to $T_0 = 0$ K since no process is observed at the starting temperature of experiment. The dependence of the conversion on temperature can be then expressed from eq. (7) as

$$\alpha = 1 - \exp\left[-\frac{A}{\beta D}\left(e^{DT}-1\right)\right] \tag{8}$$

The conversion of photooxidation was defined by the relationship

$$\alpha = \frac{c_0 - c}{c_0} \tag{9}$$

where c_0 and c are the initial and actual BaP concentrations, respectively.

The experimental dependences of conversion on temperature, shown in Tab. 1, were treated simultaneously for all heating rates applying eq. (7) by the program Origin (OriginLab, Northampton, Massachusetts, USA). The minimized parameters were A and D. The resulting values are: $A = (2.8 \pm 5.9) \times 10^{-14}$ min^{-1}, $D = (5.86 \pm 0.43) \times 10^{-2}$ K^{-1}. In Fig. 1 it can be seen that a plausible agreement between the experimental and calculated values of conversion was reached.

Knowing the kinetic parameters, modelling of the process of BaP photooxidation for any time/temperature regime can be carried out. As an example, for an isothermal process, the integration of eq. (4) leads to the result

$$\alpha = 1 - \exp\left[-A e^{DT} t\right] \tag{10}$$

Tab.1. Dependence of α on temperature during heating of benzo[*a*]pyrene in glyceryl trioctanoate at various heating rates.

T [K]	α				
	2 K·min^{-1}	1 K·min^{-1}	0.5 K·min^{-1}	0.2 K·min^{-1}	0.1 K·min^{-1}
303.15	0	0	0	0	0
313.15	0.0000	0.0000	0.0000	0.0153	0.0000
323.15	0.0200	0.0030	0.0159	0.0186	0.0001
333.15	0.0210	0.0080	0.0076	0.0228	0.0028
343.15	0.0261	0.0047	0.0085	0.0019	0.0118
353.15	0.0175	0.0184	0.0049	0.0171	0.0207
363.15	0.0180	0.0128	0.0135	0.0266	0.0289
373.15	0.0164	0.0065	0.0061	0.0159	0.0216
383.15	0.0042	0.0196	0.0099	0.0226	0.0170
393.15	0.0169	0.0096	0.0100	0.0010	0.0093
403.15	0.0172	0.0105	0.0024	0.0029	0.0108
413.15	0.0124	0.0184	0.0054	0.0024	0.0099
423.15	0.0063	0.0184	0.0209	0.0290	0.0093
433.15	0.0210	0.0184	0.0702	0.0932	0.1621
443.15	0.0231	0.0224	0.1839	0.2294	0.5730
453.15	0.0352	0.0741	0.4969	0.5935	0.8227
463.15	0.1005	0.2482	0.6997	0.8598	0.8868
473.15	0.2132	0,5000	N. M.	N. M.	N. M.

N. M. – not measured.

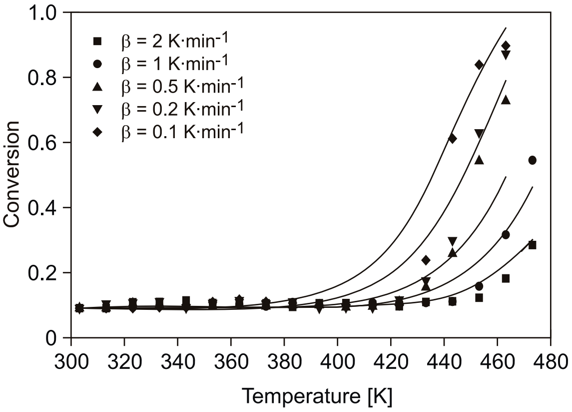

Fig. 1. Experimental (points) and optimized (lines) dependences of the conversion of benzo[*a*]pyrene photooxidation in glyceryl trioctanoate on temperature for various heating rates.

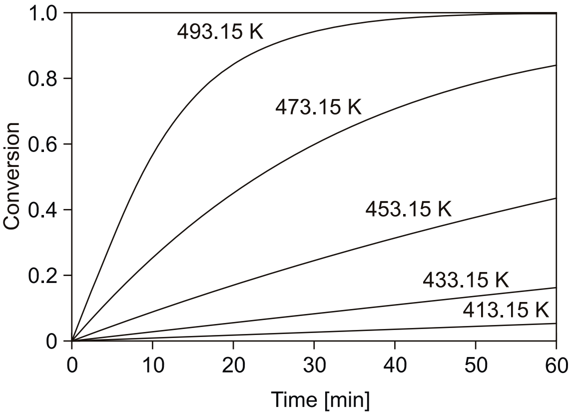

Fig. 2. Dependences of conversion of BaP photooxidation on time for various temperatures, calculated by eq. (10).

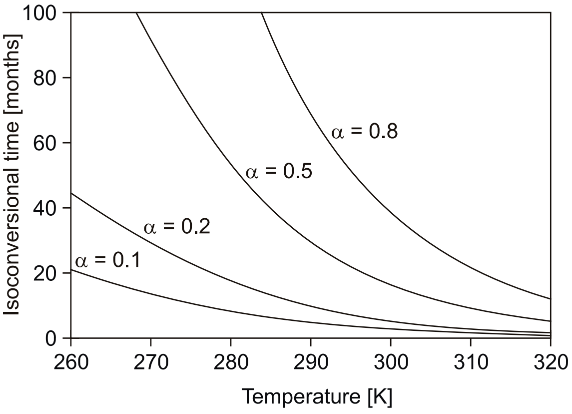

Fig. 3. Dependences of isoconversional time on constant temperature for various levels of the conversion of BaP photooxidation.

The dependences of conversion of BaP photooxidation on time for various temperatures, calculated by eq. (10) using the kinetic parameters obtained, are shown in Fig. 2. It can be seen that the conversion increases with time and also with the constant temperature. Within 1 h, a detectable decrease in BaP concentration can be attained for temperatures around 400 K. Within the given time interval, complete BaP photooxidation is attained for temperatures over 490 K.

It has been previously demonstrated [15, 16] that the temperature function given by eq. (2) provides reasonable lifetime estimations for lower temperatures in concordance with experience. From eq. (10), the relationship between the isoconversional time and temperature can be obtained:

$$t_\alpha = \frac{-\ln(1-\alpha)}{A\exp[DT]} \tag{11}$$

where t_α is the isoconversional time, i.e. the time at which the chosen value of conversion is reached for the given temperature. The dependences of isoconversional time on constant temperature for various levels of conversion are depicted in Fig. 3. For "storage" temperatures, between 260 K and 320 K, it can be seen that a detectable decrease in BaP concentration due to photooxidation takes place within months or years.

CONCLUSIONS

1. For BaP photoxidation at non-isothermal conditions with linear heating, the equations and kinetic parameters *A* and *D* characterizing the kinetics of the process were obtained.
2. The kinetic parameters *A* and *D* make possible the modelling of the process for any time/temperature regime.
3. This approach simplified considerably the complexity of the overall process taking place in the food matrix during photooxidation and enables modelling the kinetics of the process without a deeper insight into its mechanism.
4. Advantage of the multiple heating rate approach lies in the fact that a larger part of the kinetic hypersurface is mapped so that the kinetic parameters obtained are more reliable and trustworthy as when obtaining the kinetic parameters just from a single kinetic curve.

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