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STUDY OF A BIOLOGICAL OSCILLATION SYSTEM BY A MICROCALORIMETRIC METHOD

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Abstract

The power–time curves of a biological oscillation system were determined for different temperatures, acidities and carbon sources, by using a 2277 thermal activity monitor. The apparent activation energy and order of the oscillation reaction were calculated from the induction period (t_{in}) and the first oscillation period (t_p). The regularity of the biological oscillation system is discussed.

Keywords: acidity, biological oscillation, carbon source, microcalorimetric method, temperature

Introduction

Since Beloasov first reported that the homogeneous system of citric acid oxidized by bromic acid in the presence of Ce^{3+} as catalyst could produce an oscillation reaction, much research has been focussed on the oscillation regularity [1, 2], for example with amino acid [3], carbohydrate [4] and cellulose [5].

We earlier reported [6, 7] calorimetric curves of B–Z oscillation reaction systems at different temperatures, and the effects of the environmental conditions on the growth of the petroleum microbe, determined by means of microcalorimetry. In the present work, the power–time curves of a biological oscillation system were determined, for growth of the petroleum microbe in a medium with different long-chain alkanes, at different temperatures and acidities. The apparent activation energy and the order of the oscillation reaction were calculated from the induction period (t_{in}) and the first oscillation period (t_p); a non-linear relationship was established.

Experimental

Instrument

The 2277 thermal activity monitor was used in this experiment, the working temperature range of which is 10–90°C; it was maintained constant to within $\pm 2 \cdot 10^{-4}$°C at the given temperature. This system is very sensitive: the detection limit is 0.15 μW and the baseline stability (over a period of 24 h) is 0.2 μW.

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A glass electrode pH-meter was used (mode HM-20S, TOA Electronics Ltd, Japan) with a pH range of 0.00–14.00.

Materials

The petroleum microbe (bacterium K) was employed; it was obtained from Shandong University.

Medium A contained NaCl (0.5 g), $(NH_4)_2SO_4$ (0.1 g), $MgSO_4 \cdot 7H_2O$ (0.025 g), KH_2PO_4 (0.5 g), $K_2HPO_4 \cdot 3H_2O$ (1 g) and yeast extract (0.1 g) per 100 mL water; the pH of the medium was 8.60, 7.20 or 6.55.

Medium B contained 0.4 mL Tween 80 solution (2%, *v/v*) and dodecane (2%, *v/v*) per 100 mL medium A.

Medium C contained 0.4 mL Tween 80 solution (2%, *v/v*) and tetradecane (2%, *v/v*) per 100 mL medium A.

Medium D contained 0.4 mL Tween 80 solution (2%, *v/v*) and hexadecane (2%, *v/v*) per 100 mL medium A.

Tween 80 is a surfactant, which incorporates the alkanes into medium A. The media were sterilized at 120°C for 30 min.

Experimental method

The flow-through mode was used in these experiments. The flow tubing was first cleaned and sterilized, distilled water was pumped through the system for 30 min at a flow rate of 30 mL h^{-1}, 0.1 mol dm^{-3} HCl was pumped through for 30 min at a flow rate of 30 mL h^{-1}, and alcohol solution (75%) was then pumped through for 30 min at a flow rate of 30 mL h^{-1}.

Once the system had been cleaned and sterilized, sterilized distilled water was pumped through the system for 30 min at a flow rate of 10 mL h^{-1}, and the baseline was then determined. Next, the bacterial sample and the medium were pumped into the flow cell system and the monitor began to record the power–time curves under the conditions of different temperatures, acidities and carbon sources (hexadecane or tetradecane or dodecane). When the recording pen returned to the baseline and became stabilized, the process of bacterial growth was completed.

Results and discussion

Experimental results

The power–time curves of bacterium K growth were determined under the conditions of different carbon sources, acidities and temperatures. These curves are shown in Figs 1–3.

The power–time curves in Figs 1–3 show the biological oscillation phenomenon during the metabolic processes of bacterium K for the different temperatures, acidities and carbon sources.

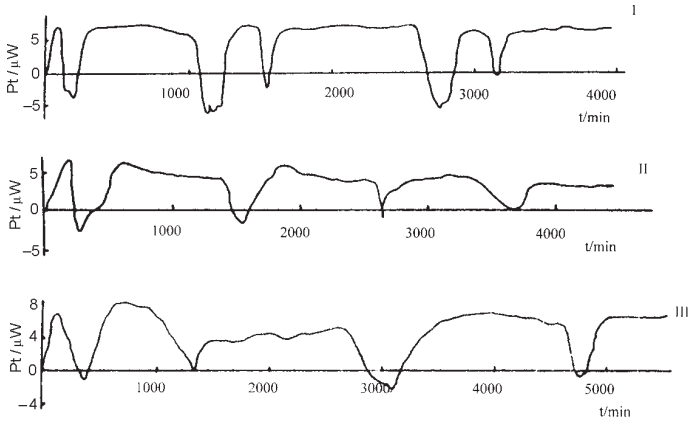

Fig. 1 Power–time curves of bacterium K in medium C (I – pH=8.60; II – pH=7.20 and III – pH=6.55) at 323 K

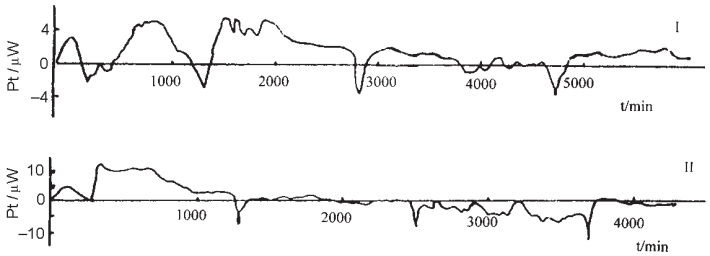

Fig. 2 Power–time curves of bacterium K in medium C at different temperatures (I – 318 K; II – 310 K) at pH=6.55

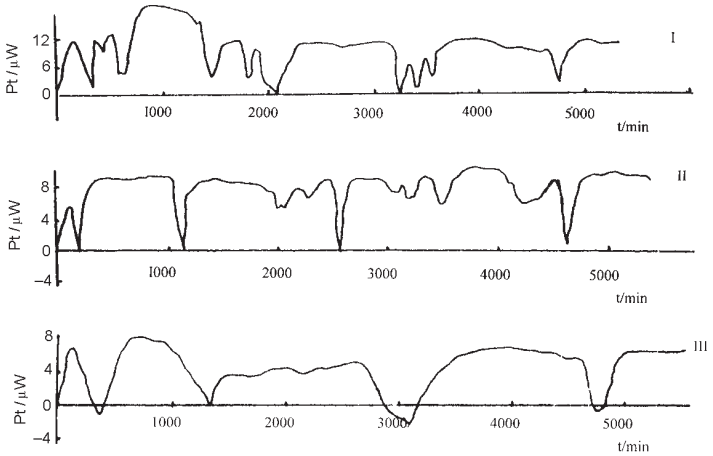

Fig. 3 Power–time curves of bacterium K in medium B, D and C, with different carbon sources (I – dodecane; II – hexadecane and III – tetradecane) at 323 K and pH=6.55

As compared with chemical oscillation, biological oscillation has the particular feature that the oscillation period changes with time, the induction period and the first oscillation period obeying a different rule. In this paper, we choose the first oscillation period.

Effects of different carbon sources on the biological oscillation system

We studied the growth of bacterium K in medium B, C and D (containing dodecane, tetradecane or hexadecane) at 323 K and pH=6.55.

The values of t_{in} and t_p were obtained from curve III in Fig. 1 and curves I and II in Fig. 3, and are listed in Table 1.

Table 1 Values of t_{in} and t_p for different carbon sources at 323 K and pH=6.55

	Dodecane	Tetradecane	Hexadecane
t_{in}/min	333	281	228
t_p/min	1122	1156	1176

From these data, the following linear equations were established:

$$t_{in}/\text{min}=648-26.25(12+2n) \quad n=0, 1, 2 \quad r=-0.9999 \tag{1}$$

$$t_p/\text{min}=962+13.5(12+2n) \quad n=0, 1, 2 \quad r=0.9890 \tag{2}$$

Effects of different temperatures on the biological oscillation system

We studied the growth of bacterium K in medium C at pH=6.55 and different temperatures. The values of t_{in} and t_p obtained from curve III in Fig. 1 and curves I and II in Fig. 2 are shown in Table 2.

Table 2 Values of t_{in} and t_p in medium C at different temperatures and pH=6.55

T/K	310	318	323
t_{in}/min	345	303	281
t_p/min	1256	1180	1156

According to a literature method [8], since $\ln(1/t)=-E/(RT)+C$, the t_{in} and t_p data were used to plot $\ln(1/t)$ *vs.* $1/T$:

$$\ln\frac{1}{t_{in}}=-0.73867-\frac{1582.4}{T} \quad r=-0.9999 \quad E_{in}=13.156 \text{ kJ mol}^{-1} \tag{3}$$

$$\ln\frac{1}{t_p}=-5.02886-\frac{652.3}{T} \quad r=0.9902 \quad E_p=5.423 \text{ kJ mol}^{-1} \tag{4}$$

Effects of different acidities on the biological oscillation system

We determined the power–time curves at different acidities in medium C at 323 K. The values of t_{in} and t_p obtained from curves I, II and III in Fig. 1 are given in Table 3.

Table 3 Values of t_{in} and t_p at different acidities in medium C at 323 K

pH	8.60	7.20	6.55
t_{in}/min	178	249	281
t_p/min	1375	1232	1156

According to Smoes [9], $t=hC_A^a C_B^b C_C^c$.... Thus, assuming that no other parameter changes except the acidity, the following equations were obtained:

$$\log t_{in}=3.0945+0.09791\log C_{H^+} \quad r=-0.9984 \tag{5}$$

$$\text{or } t_{in}=1243C_{H^+}^{0.09791}$$

$$\log t_p=2.8267-0.03633\log C_{H^+} \quad r=0.9984 \tag{6}$$

$$\text{or } t_p=670.9C_{H^+}^{0.03633}$$

The apparent activation energy and order of reaction were calculated from the values of t_{in} and t_p at different temperatures and acidities; non-linear relationships were established:

$$\frac{1}{t_{in}} \propto C_{H^+}^{-0.09791}\exp\left(-\frac{13156}{RT}\right) \tag{7}$$

$$\frac{1}{t_p} \propto C_{H^+}^{-0.03633}\exp\left(-\frac{5423}{RT}\right) \tag{8}$$

Conclusions

A biological oscillation phenomenon occurs when the petroleum microbe (or bacterium K) degrades dodecane, tetradecane and hexadecane in medium B, C and D.

From the power–time curves of the oscillation system, we can obtain the first oscillation period (t_p) and the induction period (t_{in}). For different carbon sources (dodecane, tetradecane and hexadecane), the values of t_p increase with carbon number (n) and the values of t_{in} decrease with (n); for different temperatures (310, 318 and 323 K), the values of t_{in} and t_p decrease with increasing temperature; for different acidities (pH=8.60, 7.20 and 6.55), the values of t_{in} decrease with pH, whereas the values of t_p increase with pH.

These power–time curves of the biological oscillation system allow the calculation of many data that are very useful in studies of the properties of this biological oscillation system.

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