

STUDENT VERSION

DNA Degradation in Rat Plasma

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STATEMENT

We quote the abstract from a source paper [1]:

A major obstacle in gene delivery is the transport of intact plasmid DNA (pDNA) to target sites. We sought to investigate the kinetic processes underlying the degradation of pDNA in a rat plasma model, as this is one of the main components responsible for the clearance of pDNA after intravenous administration. We further sought to construct a complete kinetic model to describe the degradation of all three topoforms (supercoiled, open circular, and linear) of pDNA in a rat plasma model. Supercoiled pDNA was incubated in isolated rat plasma at 37°C in vitro. At various time points, the plasma was assayed by electrophoresis for the amounts of supercoiled, open circular, and full-length linear pDNA remaining. The calculated amounts remaining were fit to linear differential equations describing this process. In this model, pDNA degradation is considered to be a unidirectional process, with supercoiled degrading to open circular and then to the linear topoform. The calculated kinetic parameters suggested that supercoiled pDNA degrades in rat plasma with a half-life of 1.2 minutes, open circular pDNA degrades with a half-life of 21 minutes, and linear pDNA degrades with a half-life of 11 minutes. Complexation of pDNA with liposomes resulted in a portion of the supercoiled plasmid remaining detectable through 5.5 hours.

The complete paper, [1], is available in Supporting Docs for this Modeling Scenario.

We present the system of linear differential equations model (1) and the data in Table 1 from the article [1] and ask you to explain the reasonableness of the model, to solve the model, to estimate the parameters in the model, and then to compare their model results with the data.

The model concerns three kinds of pDNA, denoted *SC* (supercoiled), *OC* (open circular), and *L* (linear) present in rats at time t in minutes and attempts to model the degradation of the pDNA,

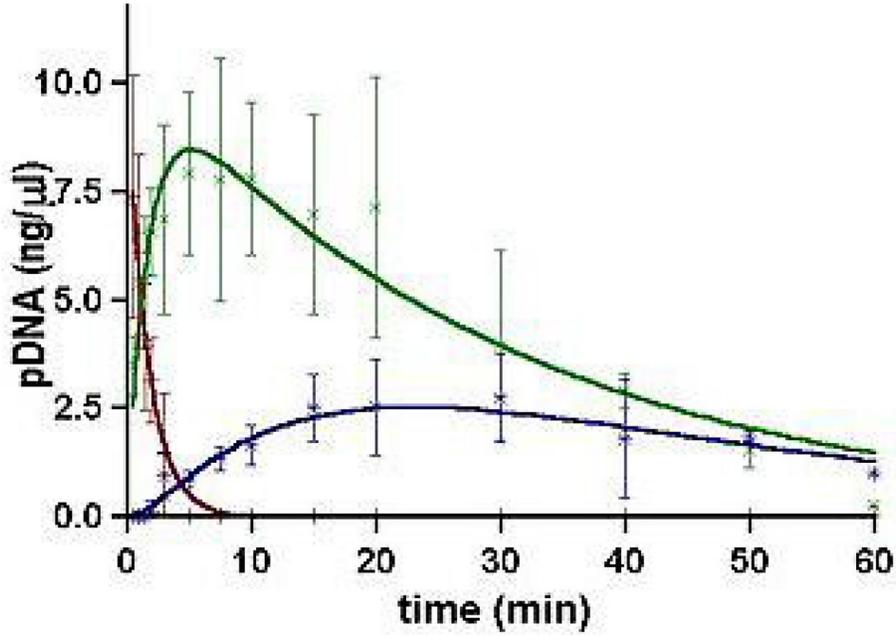


Figure 1. Plot of data and model from [1] with caption, “Experimental and fitted data based on the kinetic model described in the text. Data points represent actual experimental data. Lines represent values predicted by the model. The values are expressed as \pm SD of 6 rats. Key: + supercoiled, x open circular, * linear.”

starting at the supercoiled state, through the open circular form, until a final linear form is reached for the pDNA.

We obtained the data presented for each of the three types of pDNA on concentrations of the respective pDNA in $\text{ng}/\mu\text{l}$ by enlarging Figure 3 from the source [1]. We show the plots in Figure 1.

The Model

In [1] the following model (1) is posed for the transition between the three types of pDNA.

$$\begin{aligned}
 SC'(t) &= -k_s \cdot SC(t) \\
 OC'(t) &= k_s \cdot SC(t) - k_o \cdot OC(t) \\
 L'(t) &= k_o \cdot OC(t) - k_l L(t).
 \end{aligned} \tag{1}$$

We extracted the data (by visual estimation) in Table 1 from Figure 1 and encourage you to try your hand at such extraction to corroborate our data and/or obtain more data points.

Time (min)	Supercoiled pDNA
0	7.5
1.0	6.0
3.5	1.25
5.0	1.0
7.5	0.1

Time (min)	Open Circular pDNA
0.5	4.0
2	6.0
5	7.9
7.5	7.8
10	7.9
15	7
20	7.4
30	4.0
40	3.5
50	2.6

Time (min)	Linear pDNA
1	0
5	0.8
10	1.25
15	2.5
20	2.5
30	3.0
40	2.4
50	2.3
60	1.2

Table 1. Data on three different pDNA's extracted from Figure 3 in [1].

Activities

1. Explain the reasonableness of the mode (refeq:model).
2. Solve the model.
3. Estimate the parameters in the model.
4. Compare their model results with the data.
5. Confirm the half-life data from the Abstract above.

REFERENCES

- [1] Houk, B. E., G. Hochhaus, and J. A. Hughes. 1999. Kinetic Modeling of Plasmid DNA Degradation in Rat Plasma. *AAPS PharmSci.* 1(3): 15-20.