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A model for Zinc responses of young pigs

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A model for Zinc responses of young pigs

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Abstract

In a number of studies the feeding of pharmacological levels of Zinc to weanling pigs was examined. The publications agree in the fact that the achieved performance increase of the pigs depends on the amount of Zinc as well as on the type of mineral complex used as feed additive. The results are based on measurements of the animals' body weight and the Zinc concentration in the blood within the first two weeks of the experiments. From the measurement values, conclusions are drawn about adequate Zinc doses, but a deeper understanding of the physiological processes that influence the Zinc distribution in the organism is still missing. In this report, we want to close this gap by using a systems biology approach. We present a small mathematical model that describes the organic feedback mechanisms with the help of differential equations. The simulation results not only match with the measured values, but also give insight into non-measurable processes, which will hopefully contribute to animal experiments being organized in a more selective and reduced way.

Keywords: systems biology, ordinary differential equations, feeding study, weanling pigs

MSC: 92C42, 92C30

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1 Introduction

Many studies have documented that Zinc causes an improvement of performance of weanling pigs [14, 4]. Recent studies, however, suggest that this depends to a very great extent on the dosage and source of the added Zinc, as a contrary effect (i.e. a deterioration of the performance) occurs at high dosage [11]. The Zinc source is relevant insofar as different mineral complexes have varying bio-availabilities and thus the amounts added to the feed have to depend on the Zinc source [3]. Therefore, our objectives were to find a proper model to reproduce the (positive and negative) influence of Zinc on the performance of the weanling pigs and estimate the bio-availabilities of four zinc sources: Zinc lysine (ZnLys), zinc oxide (ZnO), zinc methionine (ZnMet) and zinc sulfate (ZnSO₄). Similar models for the Zinc metabolism of, e.g., humans [10], rats [8] or pigs [15] have already been published, but none of them included the negative influence of higher Zinc dosage. A proper mathematical tool for the purpose of modelling in the case of unknown exact mechanisms are Hill functions.

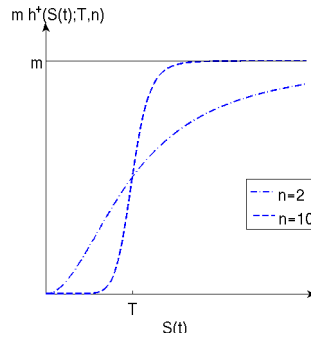


Figure 1: Positive Hill function with two different exponents.

Hill functions are used to model stimulatory or inhibitory effects,

$$H^+(S, T; n) = \frac{(S/T)^n}{1 + (S/T)^n}, \quad H^-(S, T; n) = \frac{1}{1 + (S/T)^n}.$$

Here, $S \geq 0$ denotes the influencing substance, $T \geq 0$ the threshold, and $n \geq 1$ is called the Hill coefficient. A Hill function is a sigmoidal function between zero and one that switches at the threshold $S = T$ from one level to the other with a slope specified by n , see Fig. 1.

2 Model Development

The model takes into account the data published in [14] and describes the dependencies between the following three components:

- $c(t)$: Zn plasma concentration in mg/L, $c(0) = 0.85$ mg/L
- $w(t)$: weight in kg, $w(0) = 7.5$ kg
- $i(t)$: daily feed intake in kg/d

The model is based on the following assumptions:

1. The Zn plasma concentration depends on the level of Zn supplementation, the bio-availability of Zn in the specific compound, and the blood volume (which depends on a piglet's weight).
2. The weight of a pig depends on the daily feed. The higher the feeding intake, the larger the weight gain.

3. The weight gain is stimulated if the Zn concentration in the blood is above a certain threshold (T_{c1}) and inhibited if the Zn concentration exceeds a higher second threshold (T_{c2}).
4. After a certain amount of time, $w(t)$ attains steady state with $w^{eq} = 300kg$.
5. The daily feed intake is stimulated if the weight is below a certain threshold and remains constant if the weight exceeds a second threshold.

The system of equations is as follows:

$$\frac{d}{dt}c(t) = \frac{s}{p \cdot w(t)^n} \cdot \exp(-f) \cdot i(t) - cl_{Zn} \cdot c(t) \quad (1)$$

$$\frac{d}{dt}w(t) = g \cdot i(t) \cdot (1 + w_{stim} \cdot H^+(c, T_{c1}; n_{c1}) - w_{inh} \cdot H^+(c, T_{c2}; n_{c2})) - p_{11} \cdot w(t) \quad (2)$$

$$i(t) = w_{par1} \cdot w(t) \cdot H^-(w, T_{w1}; n_{w1}) + w_{par2} \cdot H^+(w, T_{w2}; n_{w2}) \quad (3)$$

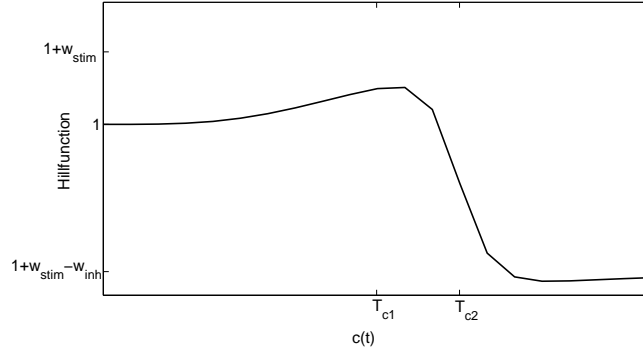


Figure 2: $1 + w_{stim} \cdot H^+(c, T_{c1}; n_{c1}) - w_{inh} \cdot H^+(c, T_{c2}; n_{c2})$ shows the desired time course.

In the following we give a detailed explanation of the equations:

- $\exp(-f)$ is the bio-availability, which is necessary as different zinc sources have different grades of availability for the organism. Bio-availability values are between 0 and 1 (i.e. 0% and 100%). If the bio-availability has the value x , it means that of a given dose y mg, $x \cdot y$ mg are disposable in the circulatory system.
- p_{11} assures that $w(t)$ reaches steady state (see next section).
- The term $1 + w_{stim} \cdot H^+(c, T_{c1}, n_{c1}) - w_{inh} \cdot H^+(c, T_{c2}, n_{c2})$ represents the stimulation/inhibition of feed caused by different zinc levels in the plasma, see Fig. 2. As described in the introduction, Hill functions are a mathematical tool for model improvement (deterioration of the performance) in case of unknown exact mechanisms.

3 Steady States

Trying to assure that $w(t)$ reaches steady state (to avoid infinite weight gain of the simulated pigs), we introduced parameter p_{11} . In the following the dependence of p_{11} on other parameters is calculated. w^{eq} and i^{eq} are the assumed steady state values of $w(t)$ and $i(t)$. It holds,

$$\frac{d}{dt}w(t) = 0 = g \cdot i^{eq} \cdot (1 + w_{stim} \cdot H^+(c, T_{c1}, n_{c1}) - w_{inh} \cdot H^+(c, T_{c2}, n_{c2})) - p_{11} \cdot w^{eq},$$

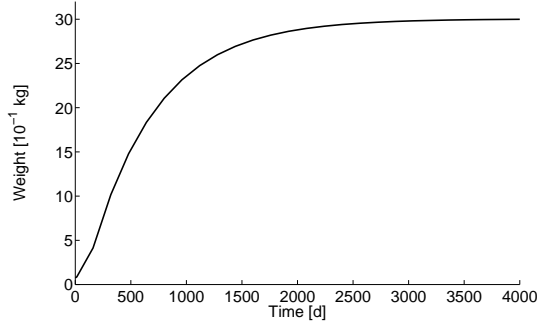


Figure 3: Simulated evolution of weight $w(t)$ over a period of 4000 days.

from which follows

$$p_{11} = \frac{g \cdot i^{eq} \cdot [1 + w_{stim} \cdot H^+(c, T_{c1}, n_{c1}) - w_{inh} \cdot H^+(c, T_{c2}, n_{c2})]}{w^{eq}}$$

Now we assume that in the long term $c(t)$ evens out at a value below parameters T_{c1} and T_{c2} , so that the Hill functions are zero. This assumption is checked below. This results in

$$p_{11} = \frac{g \cdot i^{eq} \cdot (1 + w_{stim} \cdot 0 - w_{inh} \cdot 0)}{w^{eq}} = \frac{g \cdot i^{eq}}{w^{eq}}$$

Since $c(t)$ depends on $w(t)$ and $i(t)$, $c(t)$ should also reach a steady state below parameters T_{c1} and T_{c2} , as assumed above:

$$\begin{aligned} \frac{d}{dt}c(t) = 0 &= \frac{s}{p \cdot (w^{eq})^n} \cdot \exp(-f) \cdot i^{eq} - cl_{Zn} \cdot c^{eq} \\ \Rightarrow c^{eq} &= \frac{s}{p \cdot (w^{eq})^n \cdot cl_{Zn}} \cdot \exp(-f) \cdot i^{eq} \end{aligned}$$

For ZnLys, for example, $c^{eq} = 0.0045$ which is far below T_{c1} and T_{c2} . In Fig. 5 the weight gain in a long term simulation for ZnLys shows that the desired result is achieved.

4 Data

The data points were created using data published in [14], namely the data of trial 1. In this trial the pigs were fed either a diet with no zinc added (control) or a diet with 3000mg/kg of ZnO, ZnMet, ZnLys or ZnSO4 added. The average initial weight of the pigs was 7.5kg. The data points for $w(t)$ were generated using the given average daily weight gain for week 1 and week 2. The data points for $c(t)$ and $i(t)$ are average values for week 1 and week 2 respectively.

5 Mathematical Algorithms

The main difficulty is not to simulate the system, i.e. to solve the differential equations, but to identify the unknown parameters. We will briefly describe the mathematical techniques that we use for parameter identification.

Formally, the system of differential equations can be written as

$$\mathbf{y}'(t, \mathbf{p}) = f(\mathbf{y}(t, \mathbf{p}), \mathbf{p}),$$

where $\mathbf{y}(t, \mathbf{p}) = (y_1(t, \mathbf{p}), \dots, y_n(t, \mathbf{p}))$ denotes the solution vector for a given parameter vector $\mathbf{p} = (p_1, \dots, p_q)$. Assume there are m experimental data points varying in the selected component at different time points,

$$z_k = \hat{y}_{j_k}(t_k), \quad k = 1, \dots, m, \quad j_k \in \{1, \dots, n\},$$

associated with corresponding measurement tolerances δz_k . Here $\hat{y}_{j_k}(t_k)$ denotes the measurement of component y_{j_k} at time t_k . The m to n mapping j_k assigns to every measurement time point t_k one of the n components of \mathbf{y} .

Parameter identification is equivalent to solving the least squares minimization problem

$$I(\mathbf{p}) = \mathbf{F}(\mathbf{p})^T \mathbf{F}(\mathbf{p}) \rightarrow \min_{\mathbf{p}},$$

where $\mathbf{F}(\mathbf{p}) = (F_1(\mathbf{p}), \dots, F_m(\mathbf{p}))$ is a vector of length m with entries

$$F_k(\mathbf{p}) = \frac{y_{j_k}(t_k, \mathbf{p}) - z_k}{\delta z_k}.$$

That means we want to minimize the relative deviation of model and data at the measurement time points t_k . The above problem, which is highly nonlinear in \mathbf{p} , can be solved by affine covariant Gauss-Newton iteration, see [5], where each iteration step i requires the solution of a linear least squares problem,

$$J(\mathbf{p}^i) \Delta \mathbf{p}^i = \mathbf{F}(\mathbf{p}^i).$$

The k th row of the Jacobian ($m \times q$)-matrix $J(\mathbf{p})$ has the form

$$J(\mathbf{p})(k, :) = \nabla_{\mathbf{p}} y_{j_k}(t_k, \mathbf{p}),$$

thus representing the sensitivity of the solution \mathbf{y} with respect to the parameters \mathbf{p} at the time points of measurements. An analysis of the matrix $J(\mathbf{p})$ gives some hints whether the current combination of model and data will permit an actual identification of the parameters. Parameters with very small sensitivity have nearly no influence on the solution and can therefore not be estimated. In this case the entries of the corresponding column in $J(\mathbf{p})$ (and thus the weighted l_2 column norm) are almost zero. Furthermore, some of the parameters might be linearly dependent, which leads to nearly identical columns in $J(\mathbf{p})$. In both cases the matrix $J(\mathbf{p})$ will be singular or, from a numerical point of view, nearly singular.

Linearly independent parameters can be identified by analyzing their subcondition. Let us consider the QR -decomposition of $J(\mathbf{p})$. By a suitable permutation of the matrix columns of $J(\mathbf{p})$, the diagonal elements of the upper triangular matrix R can be ordered in the form $r_{11} \geq r_{22} \geq \dots \geq r_{qq}$. The *subcondition* of parameter p_j is given by

$$sc_j = r_{11}/r_{jj}.$$

Thus, the permutation of matrix columns corresponds to a new ordering of parameters according to increasing subcondition. The subcondition indicates whether a parameter can be estimated from the given data or not. Only those parameters can be estimated for which

$$sc_j < 1/\epsilon,$$

where ϵ is the relative precision of the Jacobian $J(\mathbf{p})$ [6]. The above described techniques for solving a nonlinear least squares problem were first implemented in the software packages PARKIN [12, 7] and NLSCON [1, 5]. A renewed MATLAB-based version of this software, named POEM, which is especially adapted to parameter identification in ordinary differential equation models, has been used throughout the study.

6 Parameter Values

Table 1 below comprises the parameter values. As denoted in the last column some parameters have fixed values (taken from [14], [16] or [2]) and some were estimated using POEM.

Parameter	Value	Unit	Explanation	Parameter type
n	2	-	exponent	estimated
s	see Tab. 2	mg/kg	Zn supplement	fixed [14]
cl_{Zn}	2.4384	1/d	clearance of Zn from plasma	estimated
p	0.0435	L/kg	plasma volume linear factor	fixed [16]
f	see Tab. 2	-	bio-availability of Zn	estimated
w_{stim}	0.29679	kg/d	weight gain stimulated by Zn	estimated
T_{c1}	0.99676	mg/L	threshold of Zn for stimul. of weight gain	estimated
n_{c1}	3.45862	-	Hill exponent	estimated
w_{inh}	0.90001	kg/d	weight gain inhibited by Zn	estimated
T_{c2}	1.2992	mg/L	threshold of Zn for inhib. of weight gain	estimated
n_{c2}	22	-	Hill exponent	estimated
g	0.56	-	average gain/feed	fixed [14]
w_{par1}	1.9955	-	weight parameter	estimated
w_{par2}	0.88852	-	weight parameter	estimated
T_{w1}	3.807442	kg	first weight threshold	estimated
T_{w2}	10.951	kg	second weight threshold	estimated
n_{w1}	16.0545	-	Hill exponent	estimated
n_{w2}	5	-	Hill exponent	estimated
w^{eq}	300	kg	equilibrium value for $w(t)$	fixed [2]
f^{eq}	0.88754	kg	equilibrium value for $f(t)$	estimated

Table 1: Parameter values

7 Results

	Control	ZnO	ZnMet	ZnLys	ZnSO4
Zn supplement s (mg/kg)	111	2902	2856	2760	3293
bio-av. $\exp(-f)$	0.1710	0.0140	0.0156	0.0177	0.0147
simul. weight (kg) day 7	7.931	7.985	7.998	8.009	8.008
difference to data	+0.1510	+0.2050	+0.4280	+1.2090	+0.5780
simul. weight (kg) day 14	8.515	8.692	8.713	8.651	8.662
difference to data	-0.6650	-0.8380	-0.6070	+0.1010	-0.0280
simul. daily feed intake (kg) week 1	0.1328	0.1346	0.135	0.1355	0.1354
difference to data	+0.0128	+0.0046	+0.035	+0.0355	+0.0454
simul. daily feed intake (kg) week 2	0.1728	0.1842	0.1844	0.1845	0.1847
difference to data	-0.1372	-0.2158	-0.2456	-0.1355	-0.0953
simul. plasma Zn (mg/L) day 7	0.4173	0.903	0.9946	1.096	1.087
difference to data	-0.0727	-0.0370	-0.0454	-0.0140	-0.0130
simul. plasma Zn (mg/L) day 14	0.4814	1.066	1.176	1.278	1.271
difference to data	+0.0214	+0.0860	+0.0860	+0.0280	-0.1490

Table 2: Simulation results at day 7 and 14 of the treatment period and differences to the measurement values from Ref. [14].

8 Discussion

Fig. 6 shows that our model represents the zinc metabolism in the weanling pig fairly well. Comparison of our order of bio-availabilities (ZnO > ZnSO4 > ZnMet > ZnLys) to those in [17] (ZnSO4 > ZnMet > ZnO > ZnLys) shows that only ZnO differs in the ranking. There is only few literature

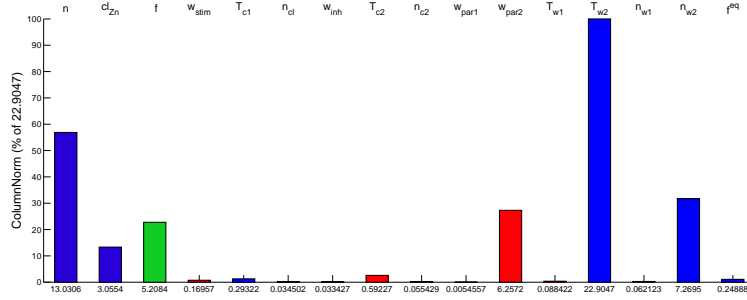


Figure 4: Column Norm of estimated parameters

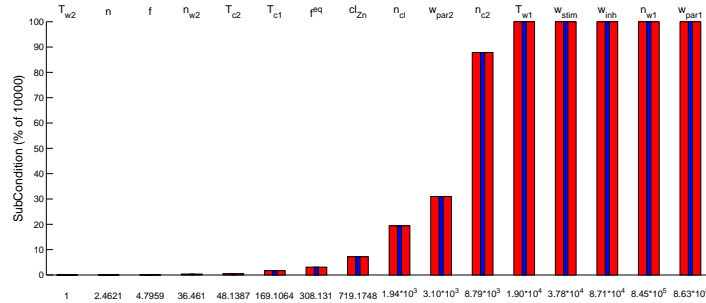


Figure 5: Subcondition of estimated parameters

about when pigs of modern breeds reach maturity. A coarse indication is given in [13]. The values listed in this publication are close to those observable in our simulated weight development, see Fig. 5.

The low number of given data points as well as the simpleness of the model pose problems. For better and more exact results, trials with more differentiated and frequent measurements are needed. Furthermore it would be necessary to insert more compartments into the model, for example liver, kidney, or excretion.

A clear weakness of our model and the estimated parameters is the underestimation of the feed intake in week 2. A reason for this may be the fact that the regulation of the appetite is a very complex process which is influenced not only by the weight and the intake of different minerals but also by genetic disposition [9]. Nevertheless a model including more organs would most likely reduce this weakness.

A further model can be found in Ref. [15]. This model has only taken into account the stimulating effect of zinc because in this trial the weanling pigs were fed marginal zinc doses. A direct comparison to this model was hardly possible since the equations are not explicitly given. Our model is a first attempt to simulate the metabolism of one single chemical element in a particular species and may be seen as a preparatory work for modelling the potassium metabolism in cattle.

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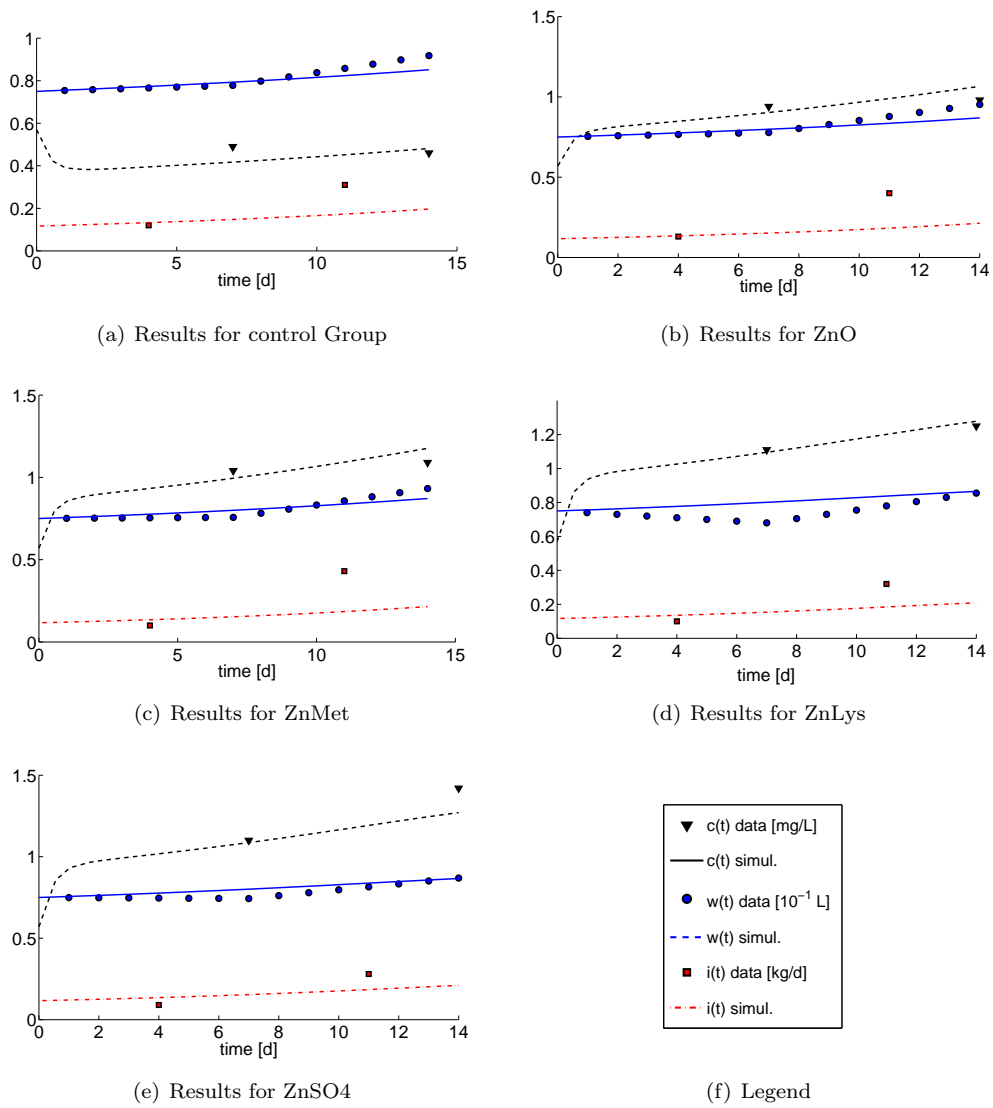


Figure 6: Simulation results and data

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