A mathematical model for the growth of elongated bones

A. Fasano[1], M.A. Herrero[2], J.M. López[3] and E. Medina[4]

[1] *Dipartimento di Matematica, Università di Firenze,*
Viale Morgagni 67/A, 50134 Firenze, Italy.
fasano@math.unifi.it.

[2] *IMI and Departamento de Matemática Aplicada, Facultad de Matemáticas,*
Universidad Complutense, Plaza de las Ciencias s/n.
28040 Madrid, Spain.
Miguel_Herrero@mat.ucm.es and director.imi@mat.ucm.es

[3] *Departamento de Morfología y Biología Celular,*
Facultad de Medicina, Universidad de Oviedo,
33006 Oviedo, Spain.
jmlopez@uniovi.es.

[4] *Departamento de Matemáticas, Universidad de Cádiz*
E11510 Puerto Real, Cádiz, Spain.
E-mail address: elena.medina@uca.es.

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Abstract

A mathematical model to describe the process of formation of bone tissue by replacement of cartilage tissue is presented and discussed. This model is based on an absorption-diffusion system which describes the interaction of two key signalling molecules. These molecules characterize the dynamics of the transition zone between the cartilage and the bone tissue.

Some experimental data are needed to estimate some model parameters. We discuss how our results are essentially unaffected by small variations, and in a particular case, not necessarily small variations in the experimental values.

1. Introduction

The ossification process for elongated bones is known as primary ossification. It starts from a pre-existing cartilage where two coupled phenomena are taking place: In a region of this cartilage, new cartilage tissue is being

produced while in other region the cartilage is being replaced by bone tissue through an invasive process. The transition region between the sheer cartilage and the advancing bone is called the Growth Plate (GP). We distinguish in the Growth Plate three zones (see figure 1).

- The Resting Zone (RZ) where the basic cells in the cartilage, named *chondrocytes*, are generated. The chondrocytes are small in size and they are irregularly arranged.
- The Proliferative Zone (PZ) where cell proliferation takes place. The chondrocytes undergo rapid divisions in such a way that the daughter chondrocytes from a cell division lie one above the other. This fact determines a formation in vertical columns and the unidirectional growth of the bone.
- The Hypertrophic Zone (HZ) where the chondrocytes enlarge in size before being overrun by the advancing bone. In this region the proliferation is arrested and the former cells enlarge in size. This increase in size determines the widening of the Growth Plate.

Fig. 1. Growth Plate: The RZ in the top, the PZ in the middle and the HZ in the bottom.

The dynamic of the Growth Plate is regulated by two proteins named Indian hedgehog (Ihh) and Parathyroid hormone-related peptide (PTHrP). The last one is produced in the RZ, then its concentration in the PZ is greater near the RZ and smaller close to the HZ (see figure 1). The PTHrP acts on proliferative chondrocytes keeping the proliferative state and inhibiting chondrocytes hypertrophy. On the other hand, Ihh is produced due to small concentrations of PTHrP and it induces the production of PTHrP. In this way a feedback loop between these two proteins is established.

This dynamics has been extensively analyzed, see for example [1–3] and some mathematical models have been proposed to describe this process, [4–7] among others. In [8] we presented a mathematical model that describes the interaction between the Ihh and the PTHrP in the PZ. The main difference with the models introduced in [4–7] is that these models depend on a large number of parameters that are not uniquely determined from the experimental data. Our model is a simpler one with a reduced number of parameters that can be estimated from the experimental results. In this work we discuss some aspects related with the model proposed and analyzed in [8]. More precisely, first we find that its solution can be determined without using the experimental value of the diffusion coefficients,

for which different values are provided in the bibliography (see for example [5], [6] or [9]). Secondly, we prove that if the experimental data undergo small variations the parameters in the model are perturbed in at most the same order of magnitude than the variations in the data.

The plan of the work is as follows. In section 2 we recall from [8] the formulation of the model. In section 3 we use the solution of the model to estimate the model parameters. As we have previously pointed out an important result of this section is that the values of the concentrations of PTHrP and Ihh can be estimated without the knowledge of the diffusion coefficients, for which different experimental values can be found in the bibliography. Next, we discuss in section 4 how the model parameters and the length of the PZ are affected by small variations in the experimental data. Moreover, in the case of a particularly small parameter (the concentration of PTHrP at the PZ/HZ boundary) we see that, not necessarily small, variations can be compensated by comparatively small variations in other model parameters in order to keep the length of the PZ constant. We finish with some conclusions in section 5.

2. The formulation of the model

We denote by $\hat{c}_i$ the concentration of Ihh and by $\hat{c}_p$ the concentration of PTHrP. We take a moving reference coordinate system in which the boundary between the PZ and the HZ is at rest. In fact, we take the space coordinate $\hat{z}$ such that this boundary is at $\hat{z} = 0$. As we shown in figure 2 we have from right to left first the RZ, then the PZ, of length Γ_P ($\hat{z} \in [0, \Gamma_P]$) and finally the HZ.

Fig. 2.   Sketch of the Ihh-PTHrP loop in a frame of reference moving with the PZ/HZ boundary.

In order to formulate the model we assume that the transport of PTHrP and Ihh occurs basically by diffusion and absorption, i.e. we suppose that the convective effects can be ignored. Then we take the steady-state linear approximation equations for a diffusion-absorption process given by the equations

(1)
$$-D_i \frac{d^2\hat{c}_i}{d\hat{z}^2} + \delta_i \hat{c}_i = 0, \qquad 0 < \hat{z} < \Gamma_P,$$

$$-D_p \frac{d^2\hat{c}_p}{d\hat{z}^2} + \delta_p \hat{c}_p = 0,$$

In order to impose boundary conditions we take into account

- PTHrP is produced at the RZ and its production is due to the presence of Ihh. Thus, we can assume that there is a flux of PTHrP at the RZ/PZ boundary which is proportional to the concentration of Ihh. Consequently, we impose that there exists a constant $\hat{\alpha}_p$ such that

$$(2) \qquad D_p \frac{d\hat{c}_p}{d\hat{z}}\bigg|_{\hat{z}=\Gamma_P} = \hat{\alpha}_p\, \hat{c}_i(\Gamma_P).$$

- At the PZ/HZ boundary we can assume that the diffusion of PTHrP is basically symmetric so that

$$(3) \qquad \frac{d\hat{c}_p}{d\hat{z}}\bigg|_{\hat{z}=0} = 0.$$

- The PZ/HZ boundary is defined by its concentration of PTHrP. Then, we can consider this concentration as a model parameter that we denoted as c_{PH} and we write

$$(4) \qquad \hat{c}_p(0) = c_{PH}.$$

- As the concentration of PTHrP at the PZ/HZ is fixed and Ihh is produced due to the low levels of PTHrP, we can assume that a constant flux of Ihh is produced when the chondrocytes leave the PZ to enter in the HZ. We denote this flux by $\hat{\gamma}_i$, then we have the condition

$$(5) \qquad -D_i \frac{d\hat{c}_i}{d\hat{z}}\bigg|_{\hat{z}=0} = \hat{\gamma}_i.$$

- The Ihh diffusion beyond $\hat{z} = \Gamma_P$ is assumed to be negligible, so that

$$(6) \qquad \frac{d\hat{c}_i}{d\hat{z}}\bigg|_{\hat{z}=\Gamma_P} = 0.$$

Summarizing we have obtained the model.

$$-D_i \frac{d^2\hat{c}_i}{d\hat{z}^2} + \delta_i\, \hat{c}_i = 0, \qquad 0 < \hat{z} < \Gamma_P,$$

$$-D_p \frac{d^2\hat{c}_p}{d\hat{z}^2} + \delta_p\, \hat{c}_p = 0,$$

$$(7) \qquad D_p \frac{d\hat{c}_p}{d\hat{z}}\bigg|_{\hat{z}=\Gamma_P} = \hat{\alpha}_p\, \hat{c}_i(\Gamma_P), \quad \frac{d\hat{c}_p}{d\hat{z}}\bigg|_{\hat{z}=0} = 0, \quad \hat{c}_p(0) = c_{PH},$$

$$-D_i \frac{d\hat{c}_i}{d\hat{z}}\bigg|_{\hat{z}=0} = \hat{\gamma}_i, \qquad \frac{d\hat{c}_i}{d\hat{z}}\bigg|_{\hat{z}=\Gamma_P} = 0.$$

3. Solution and parameter estimation

It is easy to see that the solution of (7) can be written as

(8)
$$\hat{c}_i(\hat{z}) \;=\; \frac{\hat{\gamma}_i}{D_i\,\omega_i}\,\left[\coth(\omega_i\,\Gamma_P)\,\cosh(\omega_i\,\hat{z})\,-\,\sinh(\omega_i\,\hat{z})\right],$$

$$\hat{c}_p(\hat{z}) \;=\; c_{PH}\,\cosh(\omega_p\,\hat{z}),$$

where

(9)
$$\omega_p \;=\; \sqrt{\frac{\delta_p}{D_p}}, \qquad \omega_i \;=\; \sqrt{\frac{\delta_i}{D_i}}.$$

On the other hand, the extra-boundary condition implies the model parameter constraint

(10)
$$\omega_i\omega_p\sinh(\omega_i\Gamma_P)\sinh(\omega_p\Gamma_P) \;=\; \frac{\hat{\alpha}_p\hat{\gamma}_i}{D_i\,D_p\,c_{PH}}.$$

The next step is the estimation of the model parameters from experimental data. In this sense we use the available data from ossification processes in rats. For these animals the concentration boundary values are

(11)
$$\hat{c}_i(0) \;=\; 0.5\,\mu\,M, \quad \hat{c}_i(\Gamma_P) \;=\; 0.2\,\mu\,M, \quad \hat{c}_p(\Gamma_P) \;=\; 0.3\,\mu\,M,$$

$$\hat{c}_p(0) \;=\; c_{PH} \;=\; 10^{-4}\,\mu\,M.$$

Here it is worth noting that the model parameter c_{PH} is considerably smaller than all the other boundary data. We also need the PZ length Γ_P, this data depends on the age. Experimental results are provided for three age groups, and we have

(12)
$$\begin{aligned} \Gamma_P &= 180\,\mu\,m \quad \text{for the age of 21 days,} \\ \Gamma_P &= 160.6\,\mu\,m \;\; \text{for the age of 35 days,} \\ \Gamma_P &= 137.5\,\mu\,m \;\; \text{for the age of 80 days.} \end{aligned}$$

By evaluating the second equation in (8) in $\hat{z} = \Gamma_P$ we obtain that

(13)
$$\omega_p \;=\; \sqrt{\frac{\delta_p}{D_p}} \;=\; \frac{1}{\Gamma_P}\,\cosh^{-1}\left(\frac{\hat{c}_p(\Gamma_P)}{c_{PH}}\right).$$

By setting $\hat{z} = 0$ and $\hat{z} = \Gamma_P$ in the first equation in (8) one has

$$\hat{c}_i(0) \;=\; \frac{\hat{\gamma}_i}{D_i\,\omega_i}\,\coth(\omega_i\,\Gamma_P), \quad \hat{c}_i(\Gamma_P) \;=\; \frac{\hat{\gamma}_i}{D_i\,\omega_i}\,\frac{1}{\sinh(\omega_i\,\Gamma_P)},$$

so that

$$(14) \qquad \omega_i = \sqrt{\frac{\delta_i}{D_i}} = \frac{1}{\Gamma_P} \cosh^{-1}\left(\frac{\hat{c}_i(0)}{\hat{c}_i(\Gamma_P)}\right),$$

and

$$(15) \qquad \frac{\hat{\gamma}_i}{D_i} = \frac{\hat{c}_i(\Gamma_P)}{\Gamma_P} \cosh^{-1}\left(\frac{\hat{c}_i(0)}{\hat{c}_i(\Gamma_P)}\right) \sqrt{\left(\frac{\hat{c}_i(0)}{\hat{c}_i(\Gamma_P)}\right)^2 - 1}.$$

Substitution of (15) into (10) leads us to

$$(16) \qquad \frac{\hat{\alpha}_p}{D_p} = \frac{c_{PH}}{\hat{c}_i(\Gamma_P)} \frac{1}{\Gamma_P} \cosh^{-1}\left(\frac{\hat{c}_p(\Gamma_P)}{c_{PH}}\right) \sqrt{\left(\frac{\hat{c}_p(\Gamma_P)}{c_{PH}}\right)^2 - 1}.$$

Thus, by using the experimental data (11), (12) and the formulas (13), (14), (15), (16), we determine the values of the parameters for the three age group animals. These model parameter values are shown in table 1.

Table 1.

	21 days old	35 days old	80 days old
ω_p (in μm^{-1})	0.0483	0.0542	0.0633
ω_i (in μm^{-1})	0.00870	0.00976	0.0114
$\dfrac{\hat{\gamma}_i}{D_i}$ (in $\mu M\,\mu m^{-1}$)	0.00399	0.00447	0.00522
$\dfrac{\hat{\alpha}_p}{D_p}$ (in μm^{-1})	0.0725	0.0813	0.0949

Using the data in table 1 we can plot the solution $\hat{c}_p(\hat{z})$, $\hat{c}_i(\hat{z})$ given in (8). We present the graphics for the three age groups in figure 3.

Fig. 3. Graphics of the concentrations of PTHrP (on the left) and Ihh (on the right). The three curves correspond to the three different age groups. In the case of $\hat{c}_p(\hat{z})$ the bottom graphic is for 21 days old animals, the middle for 35 days old and the top for 80 days old. On the other hand for $\hat{c}_i(\hat{z})$ we can see in the top the graphic for 21 days old, in the middle for 35 days old and in the bottom for 80 days old. The dashing lines mean the length of PZ, Γ_P, for each age.

At this point we want to remark that we have not used the values of the diffusion coefficients. However if we want to estimate the absorption coefficients δ_i and δ_p and the diffusion boundary coefficients $\hat{\gamma}_i$, $\hat{\alpha}_p$ we need

the values of the diffusivities for both molecules. In this sense we have that different values have been proposed in the literature for these parameters [5, 6,9]. Due to the fact that the two molecules have similar molecular weights we are going to take the same value for both diffusivities. More in particular we choose

$$(17) \qquad\qquad D_p \,=\, D_i \,=\, 50\,\mu m^2\,s^{-1}.$$

The model parameters corresponding to the choice (17) for our three age groups are presented in table 2 below.

Table 2.

	21 days old	35 days old	80 days old
δ_p (in s^{-1})	0.117	0.147	0.200
δ_i (in s^{-1})	0.00379	0.00476	0.00649
$\hat{\gamma}_i$ (in $\mu M\,\mu m\,s^{-1}$)	0.199	0.224	0.261
$\hat{\alpha}_p$ (in $\mu m\,s^{-1}$)	3.62	4.06	4.75

4. Robustness with respect to the experimental data

In the estimation of the model parameters we have used the experimental data (11) for the concentrations of both molecules in the boundaries of the PZ. However these values of the concentrations can undergo small variations. In this section we consider the problem of how the model parameters ω_p, ω_i, $\hat{\alpha}_p$, $\hat{\gamma}_i$ and the length of the PZ, Γ_P, are affected for small variations in the data.

In the particular case of c_{PH}, as it is significatively smaller than the other boundary concentration values, any estimate on its precise value has to be taken with caution. As a consequence it is essential to analyze the sensitivity of the model coefficients with respect to c_{PH} not only for small variations in this data.

4.1. *Dependence of the model parameters and the length of the PZ on small perturbations on the data $\hat{c}_i(0)$, $\hat{c}_i(\Gamma_P)$, $\hat{c}_p(\Gamma_P)$*

We assume that the concentration boundary values $\hat{c}_i(0)$, $\hat{c}_i(\Gamma_P)$ or $\hat{c}_P(\Gamma_P)$ undergo small variations and write the new concentration values as

(18)
$$\bar{\hat{c}}_p(\overline{\Gamma}_P) \,=\, \hat{c}_p(\overline{\Gamma}_P)\,(1+\eta_p), \quad \bar{\hat{c}}_i(0) \,=\, \hat{c}_i(0)\,(1+\eta_{i,0}), \quad \bar{\hat{c}}_i(\overline{\Gamma}_P) \,=\, \hat{c}_i(\overline{\Gamma}_P)\,(1+\eta_{i,1}).$$

Let us denote the new model parameters and the new length of the PZ as
(19)
$$\overline{\omega}_p = \omega_p\,(1 + \epsilon_p), \quad \overline{\omega}_i = \omega_i\,(1 + \epsilon_i), \quad \overline{\hat{\alpha}}_p = \hat{\alpha}_p\,(1 + \nu_p), \quad \overline{\hat{\gamma}}_i = \hat{\gamma}_i\,(1 + \nu_i),$$

$$\overline{\Gamma}_P = \Gamma_P\,(1 + \xi).$$

If we leave c_{PH} unperturbed and assume that the diffusion coefficients experiment no changes, the equations (13)-(16) transform into

$$\overline{\omega}_p\,\overline{\Gamma}_P = \cosh^{-1}\left(\frac{\overline{\hat{c}}_p(\overline{\Gamma}_P)}{c_{PH}}\right), \quad \overline{\omega}_i\,\overline{\Gamma}_P = \cosh^{-1}\left(\frac{\overline{\hat{c}}_i(0)}{\overline{\hat{c}}_i(\overline{\Gamma}_P)}\right),$$

(20) $$\frac{\overline{\hat{\gamma}}_i}{D_i}\,\overline{\Gamma}_P = \overline{\hat{c}}_i(\overline{\Gamma}_P)\cosh^{-1}\left(\frac{\overline{\hat{c}}_i(0)}{\overline{\hat{c}}_i(\overline{\Gamma}_P)}\right)\sqrt{\left(\frac{\overline{\hat{c}}_i(0)}{\overline{\hat{c}}_i(\overline{\Gamma}_P)}\right)^2 - 1},$$

$$\frac{\overline{\hat{\alpha}}_p}{c_{PH}\,D_p}\,\overline{\Gamma}_P = \frac{1}{\overline{\hat{c}}_i(\overline{\Gamma}_P)}\,\cosh^{-1}\left(\frac{\overline{\hat{c}}_p(\overline{\Gamma}_P)}{c_{PH}}\right)\sqrt{\left(\frac{\overline{\hat{c}}_p(\overline{\Gamma}_P)}{c_{PH}}\right)^2 - 1}.$$

Now by performing in (20) Taylor expansions to the first order in the perturbations η_p, $\eta_{i,0}$, $\eta_{i,1}$, ϵ_p, ϵ_i, ν_i, ν_p, ξ and using the equations (13)-(16) and the experimental data (11) we obtain that the perturbations in the experimental data and the induced perturbations in the model parameters are related through the system

$$\epsilon_p + \xi = 0.115\,\eta_p,$$

$$\epsilon_i + \xi = 0.696\,(\eta_{i,0} - \eta_{i,1}),$$

(21)

$$\nu_i + \xi = 1.89\,\eta_{i,0} - 0.887\,\eta_{i,1},$$

$$\nu_p + \xi = -\,\eta_{i,1} + 1.11\,\eta_p.$$

Note that (21) has been obtained independently of the values of the diffusion coefficients D_i and D_p and of the length of the PZ, Γ_P, (i.e. independently of the age).

If we want, for example, to keep the flux coefficient $\hat{\alpha}_p$ unperturbed, i.e.

$\nu_p = 0$, from (21) we obtain for the other model parameter perturbations

$$\xi = 1.11\,\eta_p - \eta_{i,1},$$

(22)
$$\epsilon_p = \eta_{i,1} - \eta_p,$$

$$\epsilon_i = 0.696\,\eta_{i,0} - 0.304\,\eta_{i,1} - 1.11\,\eta_p,$$

$$\nu_i = 1.89\,\eta_{i,0} + 0.113\,\eta_{i,1} - \eta 1.11\,\eta_p.$$

Thus, in order to keep the flux coefficient $\hat{\alpha}_p$ constant, a small variation in $\hat{c}_i(0)$ ($\eta_{i,0}$) implies small modifications in ω_i (ϵ_i depends on $\eta_{i,0}$) and $\hat{\gamma}_i$ (ν_i depends also on $\eta_{i,0}$), but leaves unperturbed the length of the PZ and ω_p (ξ and ϵ_p are independent of $\eta_{i,0}$).

Similar results are obtained when other model parameters are chosen to remain unperturbed. Finally, from (9) we have that $\delta_i = D_i\,\omega_i^2$ and $\delta_p = D_p\,\omega_p^2$, so that, up to the first order, the perturbations in the absorption coefficients are given by

(23)
$$\overline{\delta}_i = \delta_i\,(1 + 2\,\epsilon_i), \quad \overline{\delta}_p = \delta_p\,(1 + 2\,\epsilon_p).$$

Thus we can conclude

Proposition 4.1. *Small variations in the experimental boundary concentration values $\hat{c}_i(0)$, $\hat{c}_i(\Gamma_P)$ or $\hat{c}_p(\Gamma_P)$ induce also small variations, at most of the same order of magnitude in the length of the PZ, Γ_P, and in the model parameters in (7) δ_i, δ_p, $\hat{\gamma}_i$ and $\hat{\alpha}_p$. Moreover, this property is independent of the precise values of the diffusion coefficients D_i and D_p and of the length of the PZ, Γ_P, and consequently of the age.*

4.2. *Dependence of the absorption coefficient δ_p and the flux coefficient $\hat{\alpha}_p$ on the boundary concentration c_{PH}*

According to (11), the model parameter c_{PH} is significatively smaller than the other boundary concentration values. As a consequence it is essential to analyze the sensitivity of the model coefficients with respect to c_{PH}, not only with respect to small changes, as we have considered in the previous subsection for the other boundary concentration values, but also for considerable changes in the value of c_{PH}. We are going to analyze how the model parameters have to change in order to keep the length of the PZ constant when c_{PH} changes in a range of two orders of magnitude. More in particular, taking into account that (see (11)) the experimental value for c_{PH} is $10^{-4}\,\mu\,M$, we are going to consider values

(24)
$$c_{PH} \in (10^{-5}\,\mu\,M, 10^{-3}\,\mu\,M).$$

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From (14) and (15) it is clear that the absorption coefficient δ_i and the flux coefficient $\hat{\gamma}_i$ are independent of the value of c_{PH}. On the other hand from (13) and (16) we have that

(25)
$$\delta_p = \frac{D_p}{\Gamma_P^2} \left(\cosh^{-1} \left(\frac{\hat{c}_p(\Gamma_P)}{c_{PH}} \right) \right)^2,$$

$$\hat{\alpha}_p = \frac{D_p \, c_{PH}}{\Gamma_P \, \hat{c}_i(\Gamma_P)} \cosh^{-1} \left(\frac{\hat{c}_p(\Gamma_P)}{c_{PH}} \right) \sqrt{\left(\frac{\hat{c}_p(\Gamma_P)}{c_{PH}} \right)^2 - 1}.$$

Now, we use in (25) the values of the concentrations $\hat{c}_i(\Gamma_P)$, $\hat{c}_p(\Gamma_P)$ in (11) and the value of the diffusion coefficient D_p in (17) and plot the graphics of the model parameters δ_p and $\hat{\alpha}_p$ as functions of the concentration c_{PH} in figures 4 and 5 respectively.

Fig. 4. δ_p as a function of c_{PH} for 21 (bottom), 35 (middle) and 80 (top) days of age, respectively. Dots correspond to the experimental data $c_{PH} = 10^{-4} \, \mu M$.

Fig. 5. $\hat{\alpha}_p$ as a function of c_{PH} for 21 (bottom), 35 (middle) and 80 (top) days of age, respectively. Dots correspond to the experimental data $c_{PH} = 10^{-4} \, \mu M$.

From this figures we can see that in the range of values of c_{PH} we are interested, both model parameters are decreasing functions of the boundary concentration value c_{PH}. They take values in the ranges

(26)
$$\delta_p \in \begin{cases} (0.0631, \, 0.187) & \text{for 21 days of age,} \\ (0.0793, \, 0.235) & \text{for 35 days of age,} \\ (0.108, \, 0.320) & \text{for 80 days of age.} \end{cases}$$

(27)
$$\hat{\alpha}_p \in \begin{cases} (2.67, \, 4.58) & \text{for 21 days of age,} \\ (2.99, \, 5.14) & \text{for 35 days of age,} \\ (3.49, \, 6.00) & \text{for 80 days of age.} \end{cases}$$

From the figures and the equations (26) and (27) we conclude that *in order to keep the length of the PZ constant, considerable variations of the boundary concentration c_{PH} can be compensated with comparatively small variations in the model parameters δ_p and $\hat{\alpha}_p$*. It is also worth noticing that according to (25) both model parameters δ_p and $\hat{\alpha}_p$ are proportional to the diffusion coefficient D_p. Then, a different choice of the value of D_p implies different values of δ_p and $\hat{\alpha}_p$, but the same form of their graphics, so that our conclusion of the little sensitivity of the model to the precise value of c_{PH} is independent of the value of D_p.

5. Conclusions

We have presented a mathematical model for the Ihh/PTHrP loop that determines the growth of the PZ in primary ossification. This model is a simple one in the sense that all the equations are linear and it works with a reduced number of parameters. We have estimated the model parameters from some experimental data and we have analyzed:

- How small perturbations in the values of both concentrations at the RZ/PZ boundary and of the concentration of Ihh in the PZ/HZ boundary imply also small variations in the model parameters (absorption coefficients and boundary flux coefficients) and in the length of the PZ.

- How not necessarily small modifications in the model parameter c_{PH} (concentration of PTHrP at the PZ/HZ boundary), which is significatively smaller than the other boundary concentration values, can be compensated by means of comparatively small changes in other model parameters, the absorption coefficient of PTHrP, δ_p, and the flux coefficient of PTHrP in the RZ/PZ boundary, $\hat{\alpha}_p$, in order to keep the length of the PZ constant.

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