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A Mechanism of crystallization process of hemoglobin S

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Abstract. It has been studied as the qualitative consequence of a proposed mechanism of reaction for the formation of HbS molecular aggregations, as well as the mathematical model associated with it, for the particular case that the process of crystallization takes place at partial pressure and constant temperature.

It is showed that the results mentioned in the study can be used to explain experimental existing data, although this data are small to decide between various options that are compatible with existing experimental models and experimental data.

Also, qualitative consequences of to mechanism of reaction suggested for the formation of HbS molecular aggregations, as well as the mathematical model associated, are studied for a particular case in which to process of crystallization under constant oxygen, partial pressure and constant temperatures.

The results of such study, which are able to explain the existing experimental data, are shown. However, this is latter very scarce for being able to decide among various existing choices compatible with the model and experimental data.

1. Introduction

Sickle-cell anemia is a molecular disease which remains worldwide as a health problem [1]. The genetic origin of this disease is related to an alteration in the hemoglobin's

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amino-acids chain, leading to polymerization of Hemoglobin S (HbS), under low oxygenation conditions, deformation of red blood cells, changes in permeability and elasticity of their membranes and modifications in blood rheology and several physiological processes. The polymerization of deoxygenated HbS within the erythrocyte is influenced by several factors. The molecular mechanisms of the polymer formation play a central role in the interpretation of the pathophysiology of the disease and in the selection of the therapeutic strategies to follow up [1, 2]. Different mechanisms and models of the formation of polymers and HbS molecular dominions have been proposed, which allow the description of numerous phenomena in the literature [2]–[11].

A mechanism and a model of the polymerization, when assuming defective structural units are formed in, were presented in [3]. The model describes the influence of the concentration of HbS in the polymerization process, in absence of crystallization, and the role of other Hemoglobin, including HbS itself, whenever its three-dimensional structure or contact points are modified. As a continuation of the work done in [3], this paper is focused on the analysis of the crystallization process.

In this paper is pointed out that if in one moment there is not molecular dominions of deoxyHbS then this will not be present until a result of a polymerization process without crystallization the concentration of deoxyHbS in microtubule reach the top (β_1).

The system of equations that describe the process of crystallization–polymerization of HbS is, see [3], as

$$x(t) + ny(t) + nz(t) + w(p) = N, \tag{1}$$

$$\frac{dx}{dt} = -nP(x) - (c-d)\,x + nby, \tag{2}$$

$$\frac{dy}{dt} = -\left(b + e - f\right) y + P\left(x\right), \tag{3}$$

$$\frac{dz}{dt} = \left(\frac{c-d}{n}\right) x + (e-f)\,y, \tag{4}$$

where

$x(t)$: [deoxyHbS] in monomer states or forming part of defective structural units; $ny(t)$: [deoxyHbS] in microtubule; $nz(t)$: [deoxyHbS] in dominions; $w(p)$: [oxyHbS], depends on the p in blood; N : total [HbS]. a_i $(i = 1, \ldots, 2n)$, b, c, d, and e, are the reaction coefficients: a polymerization, b depolymerization, c crystallization by the addition of deoxyHbS monomers, d decrystallization by separation of monomers, e of crystallization by addition of microtubule; and f of the decrystallization by the separation of microtu-

bule. $n\alpha$ number of molecules of HbS that form the structural unit of the microtubule. Generally we consider $\alpha = 2$ so $a = 2 + \delta$ where $|\delta| < 1$ [2]. Finally, $P(x) = \sum_{i=1}^{2n} a_i x^i$ is the Polymerization Function.

2. Method

2.1. Beginning of the crystallization

Crystallization is a slower process than polymerization in the absence of crystallization [2, 3]. On the other hand, in initial moment of crystallization the situation in not very different that when no dominion exist and the polymerization dominions with respect the depolymerization is dominant. So that for $t \ll 1$, when crystallization begin, is possible to approximate the system of equation (1)-(4) for the system

$$\frac{dy}{dt} = -P(x), \qquad \frac{dx}{dt} = -nP(x), \qquad \frac{dz}{dt} = 0. \tag{5}$$

In the plane $z(t) = C$, the invariant region of the system, the solutions of this systems are lines with slope $-1/n$, result that is similar to the one obtained in [3]. For the case of complete absent of crystallization, those solutions can be written in the following form:

$$x + ny = C.$$

2.2. Advance crystallization

If the conditions are in favor of the crystallization process at the end of sufficiently long time, the major part of deoxyHbS will be found made part of the dominions. So that, when $t \gg 1$ (crystallization is very advanced), the system (1)-(4) can be described by means of the system

$$\frac{dx}{dt} = -(c-d)x, \tag{6}$$

$$\frac{dy}{dt} = -(e-f)y, \tag{7}$$

$$\frac{dz}{dt} = \left(\frac{c-d}{n}\right)x + (e-f)y, \tag{8}$$

and the equation of conservation of mass (1).

Finally, for some cases we should use the system of equation (1)-(4) in its general form. In the following, the fundamentals properties of the solution of those system of equation will

be discussed and the interpretably phenomenon, to prove if the mentioned mechanism of reaction is capable to explain the fundamental results.

3. Results and discussion

3.1. Beginning of the crystallization

The system of equations (1) y (5) describes the polymerization process when there are depreciable depolymerization processes at the beginning of the crystallization process, and with one total concentration effectiveness of HbS given for $N' = N - nz(t)$. If $z(t) \ll 1$ then $N' \approx N$, the polymerization process is much faster than the crystallization one, and then the effects of the time when the polymerization occurs the concentration of the HbS in the dominions is practically constant.

On the other hand, the system of equations (1) and (5)-(7) describes the process of polymerization in the absence of crystallization, see [3], with an total concentration effectiveness of HbS given for $N' = N - nz(t)$, and where the polymerization is ignored. This reflects the fact that if $z(t) \ll 1$ the process of polymerization is much more faster than the crystallization and "interpret" to the concentration of HbS in domain as a constant in the short period of time in which occurs the polymerization.

That time when the polymerization is absent, the crystallization will increment notably with small decrements of total concentration of HbS, and at the time Δt when begins the crystallization $N' = N - nz(t)$, the decrement will be sufficient from the polymerization time and crystallization be comparable and the approximation above $N' \approx N$ will occur without certain [3].

Although as the time the polymerization is absent in crystallization, equation (3) increases notably with small decreases in the total concentration of the HbS at the end of t period, when begins the process of crystallization $N' = N - nz(t)$ which will begin to decrease sufficiently in such a way that the polymerization time becomes comparable to the crystallization and this approximation.

3.2. Advance crystallization

The system of equations (6)–(8) describes the process of the advance crystallization in absence of the polymerization, where the dominions are increscent of the monomers of desoxiHbS and increscent of microtubule units. This mechanism corresponds to the

process of homogeneous and heterogeneous molecular agglutination described in [2].

Great advance crystallization

The system of equations (8)-(10) belongs to a process of crystallization in the absence of polymerization in which the domain of deoxyHbS increases by the action of two independent mechanisms: by the addition of monomers of deoxyHbS (homogeneous processes) or by the addition of structural units of microtubule (heterogeneous processes) [2].

Integrating (6) and (7) we obtain that

$$x(t) = x_0 e^{-(c-d)t}, \;\; y(t) = y_0 e^{-(e-f)t}, \tag{9}$$

where x_0 and y_0 are the initial concentrations. Also, integrating (8) is obtained that

$$z(t) = \frac{x_0}{n} e^{-(c-d)t} - y_0 e^{-(e-f)t} + D., \quad D = \frac{x_0}{n} + y_0 + z_0, \tag{10}$$

where $z_0 =?$. For this case it can be said that $c > d$ and $e > f$ because in the other case exist a moment t_1 so that for $t > t_1$ we obtain that $z(t) < 0$, which is impossible.

There exists an interesting and particular case of (10). In the discussion before was considered that the crystallization is very advanced, when the polymerization time is highly increased (for the continuous decrease of the effective total concentration of HbS given for $N' = N - nz(t)$) that is much bigger than the crystallization time, and therefore the crystallization process will occur before the equilibrium between HbS monomers and microtubule exists. It can happen before it has arrived to the stage of very advanced crystallization, the monomers concentration has diminished to value smaller than α and therefore the deoxyHbS concentration in microtubule is virtually null ($y(\tau) = 0$). In this case, during the very advanced crystallization process, the domains will grow only to coast of the addition of deoxyHbS monomers and the expression (12) will be described like

$$z(t) = -\frac{x_0}{n} e^{-(c-d)t} + D. \tag{11}$$

3.3. Crystallization in intermediate stage

To study in detail the crystallization and polymerization processes in this stage, it is indispensable to solve the system of equations (1)-(4), that is not possible to integrate explicitly.

Nevertheless, there exists the particularly attractive case for which $z(t)$ can be known in an explicit way. If it happens, the two possible crystallization mechanisms are equally possible, that is to say if $k = c - d = e - f$, then the equation (4) can be described like

$$\frac{dz}{dt} = \left(\frac{k}{n}\right)(x + ny). \tag{12}$$

Clearing the term $x + ny$ from (1) and substituting it in (12) we obtain that

$$n\frac{dz}{dt} = -k[nz - (N - w(p))]. \tag{13}$$

In this case, the solution of the system (2)–(3) has the form

$$x(t) + ny(t) = D, \tag{14}$$

$$z(t) = \frac{1}{n}\left[(N - w(p))\left(1 - e^{-kt}\right)\right]. \tag{15}$$

Result perfectly compatible with those expressed for the formulas (10) and (11) when the condition (12) is satisfied.

Making use of general system we obtain Pfaff's equation, see [12],

$$dx + ndy + ndz = 0, \tag{16}$$

whose integral is

$$x(t) + ny(t) + nz(t) = D, \qquad D = cte. \tag{17}$$

Thus, we have obtained the first integral of system (2)-(4) that allow us to reduce it to the new system o equations

$$\frac{dx}{dt} = nP(x) - (c - d)x + nby, \tag{18}$$

$$\frac{dy}{dt} = -(b + e - f)y - P(x), \tag{19}$$

in the plane $x + ny + nz = D$, which represents an invariant region for such system.

If $e - f = c - d$, the system of equations (18)-(19) can be reduced to an equation on separable variables whose integral is

$$y = \left(-\frac{1}{n}\right)[x - ke], \qquad k = cte. \tag{20}$$

However, the last one does not allow one to decide if there really exist two mechanisms for the increasing of the deoxyHbS domains, and we can not determine which is the most important in each stage of the crystallization process.

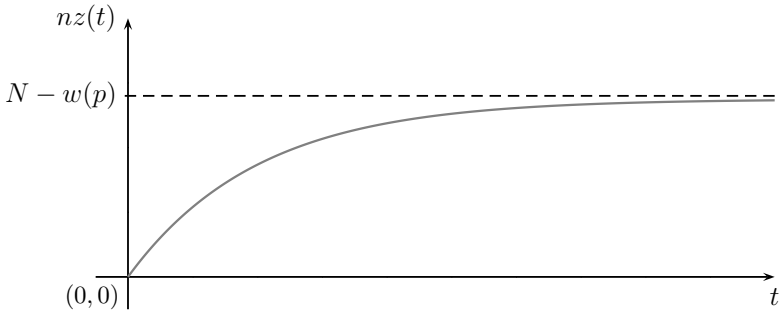

Figure 1.

Conclusions

It has been studied that the qualitative consequences of the mechanism reaction for the formation of HbS units molecules and the mathematical model associated with it in the particular case where occur to crystallization process at to partial pressure and constant temperature.

From the discussion it follows that two compatible situations can be given with the model and with the experimental existing dates:

1. That the mechanisms to increase the deoxyHbS by the addition of monomers or the addition of microtubule are equivalent, and therefore the temporal dependency of $z(t)$ is given by the formulas (20) for all t.

2. That the mentioned mechanism are not equivalent. Therefore, it is expected that at the beginning at crystallization (perhaps to few minute) predominates the increase of the domains by the addition of microtubule. In the advanced crystallization stage predominates the increase of the domains by the addition of monomers of deoxyHbS, formula (11), or both mechanism are present but independently, formula (10).

On the other hand, we can say few about the intermediate stage of crystallization in this case. At last, it is necessary to study the descriptive and predictive power of the proposed model from the quantitative point of view. Experimental methods and theoretical description of the polymerization of the HbS are still being developed.

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