



## ELECTROSTATIC POTENTIAL AND ITS ROLE IN HYDROXYAPATITE PRECIPITATION

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Electrostatic potential has been calculated for the different faces of the hydroxyapatite crystal, based on a mathematical model and using computational design software. Electrostatic potential values indicate the possibility to influence the crystalline habitus by modifying the composition of the crystallization environment. The goal of the present study is to assess the electrostatic potential at different distances from the crystal faces, and its role at the crystal-solution interface.

### INTRODUCTION

Medicine used calcium phosphate ceramics operate as replacements for human bones, as biomaterials with growing clinical applicability in the last decade. Bone substitute materials take a primordial role in current research, and represent a reliable and viable alternative for commonly used grafts which involve sampling trauma, biological hazards and aspects of physical integrity.

Bone grafts come in second place on the list of the most transplanted medical products, after blood transfusion. Recent medical achievements<sup>1</sup> strengthen the path towards bone reconstruction using synthetic materials, and there is a constantly growing demand for such new products in the treatment of bone fractures, arthritis, osteoporosis, tumors, bone infections and even spine surgery.

Most frequently used materials include a mineral fraction based on hydroxyapatite and calcium phosphates, colonized with stem cells, natural growing factors and morphogenetic bone proteins. In this respect, hydroxyapatite is very interesting,<sup>2</sup> and its interaction with other in vivo compounds (cations like  $Mg^{2+}$ , tensioactive substances) is subject to recent investigations.

### METHODS

A mathematical model has been designed for the hydroxyapatite crystalline net, one that best describes spatial models for the constituent atoms of  $Ca_{10}(PO_4)_6(OH)_2$ .

Both hydroxyapatite and fluorapatite have similar structures. Both crystallize in hexagonal system,  $P6_3/m$  symmetry group, and elementary cell parameters differences are negligible.<sup>3,4</sup>

Taking account of this, the fluorapatite is the best structure for designing a model to study the electrostatic field near the crystal surface, as it has fewer atoms per elementary cell. Moreover, the crystalline structures present high structural similarity, and the electric charges are identically distributed in space.

With this in mind, computational design procedures were started, in order to obtain a simplified mathematical model with representatively and predictive power.

Molecular formula of fluorapatite:  $Ca_5(PO_4)_3F$ . Crystallizes in hexagonal or monoclinic system,  $P6_3/m$  or  $2/m$  symmetry group. Elementary cell dimensions for  $P6_3/m$  symmetry group:  $a = b = 9.3973$ ,  $c = 6.8782$ ,  $Z = 2$ .

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The starting structure is shown in Fig. 1. Original source was a .cif file from the American Mineralogist.<sup>5</sup> Visualization program Mercury<sup>6</sup> presents, as shown in Fig. 1a, the primary structure, and in Fig. 1b the crystalline net.

Extended crystalline net generation leads to structure from Fig. 2.

One can observe the rhomboidal structure of the elementary cell, the specific element of hexagonal crystallization system.

In order to study the electrostatic potential variations at the fluorapatite crystal surface, a section of the crystalline net (Fig. 2) was generated with the Mercury software, with enough atoms in it for the computational model to be relevant and representative.

This structure was imported in the molecular modeling software HyperChem,<sup>7</sup> where the implemented computational methods present the possibility to determine and work with atomic distances and electric charges, used for the calculation of electrostatic potential values in different reference points at the crystalline surface.

Because the obtained raw structure still had superposed atoms (from the crystalline net construction) they have been removed using the script facility of the software (the script represents

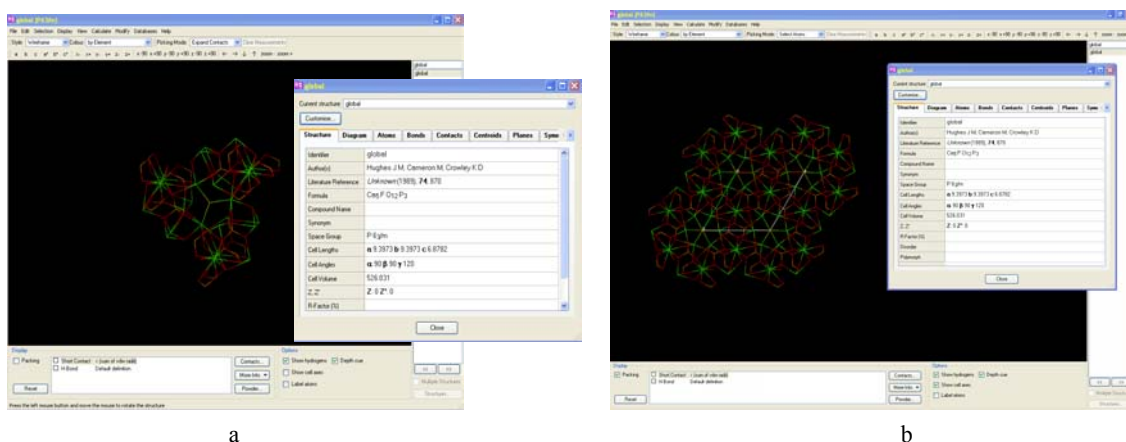
a succession of command lines, much like the structure of a computer program). The scripts were also used later, to read information from the analyzed structures.

The growing speed of crystals toward perpendicular direction on different crystal faces, and also the surface tension, will depend on the electrostatic field in the vicinity of the respective face.<sup>8</sup>

The electrostatic potential calculation was achieved by considering a reference point outside the crystal (i.e. the center of a reference atom), and summing the electrostatic influences of all the other atoms in the reference point, taking in account their electric charge and the interatomic distances between the pairs of atoms, by the formula:<sup>9</sup>

$$\vec{V}(\vec{R}) = \frac{1}{4\pi\epsilon_0} \sum_i \frac{q_i}{|\vec{R}_i - \vec{r}|}, \quad (1)$$

where:  $q_i$  = electric charge of the  $i$  ion;  $\vec{R}_i$  = position vector of  $i$  ion in the crystal;  $\vec{r}$  = position vector of the reference point where the potential is calculated;  $4\pi\epsilon_0$  = dielectric constant of the environment.



a

b

Fig. 1 – Fluorapatite: a) primary structure; b) crystalline net.

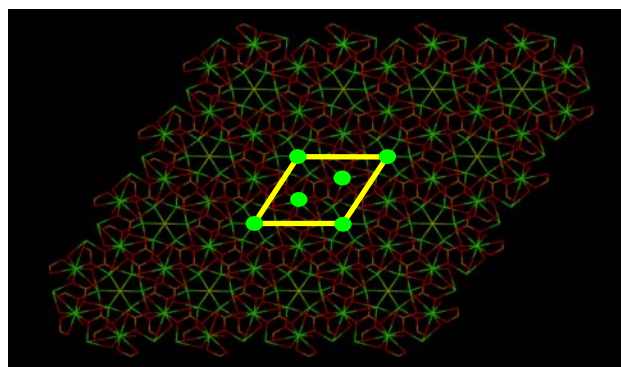


Fig. 2 – Fluorapatite, extended crystalline net considered for the study.

For this case  $\vec{r} = 0$  because the influence of the electric field is calculated right in the reference point. Also, to simplify the calculation, the influence of the dielectric constant can be neglected, assuming the effect is proportional for all faces. Besides, we are not interested in absolute values of the electrostatic potential in a certain point, but rather in relative values for comparison purposes, and discussions concerning its influence on the crystalline habitus. We then come to the simplified formula:

$$V_i = \sum_i \frac{q_i}{d_i}, \quad (2)$$

where  $q_i$  has been considered as ionic charge in classical sense, and the distance  $d_i$  between the net ions and reference points has been calculated with scripts, from system coordinates.

Below, an example of atomic cartesian coordinates (Å):

coordinates(1, 1) = -17.589, 14.305, 12.037

coordinates(2, 1) = -18.795, 16.277, 12.037

coordinates(3, 1) = -16.79, 14.181, 9.832 .....

The reference atom has been positioned in a plan that is parallel with the crystalline face, at a distance of two Van der Waals calcium radii. The

$\text{Ca}^{2+}$  Van der Waals radius is 1.05 Å, so the distance between the reference point and the crystal face was set to 2.1 Å (Fig. 3).

Inside this reference plan, the reference atom was placed in different points, and then the electric potential was computed, as a sum of all electric influences. An arithmetic mean of these calculations was considered as a comparative value for the respective crystal face, at a preset distance.

The width (or depth) of the crystalline face was set to be the size of one elementary crystalline cell, considering that atoms positioned at greater distances, inside the crystal, will exert a negligible influence on the reference point due to increased distance and the shielding effect.

A fragment of the constructed script is presented below:

```
omsgs-to-file results.txt
errors-are-omsgs
select-none
query-value atom-charge 1,1
select-atom 1,125
select-atom 1,1
name-selection selected
named-selection-value selected ? .....
```

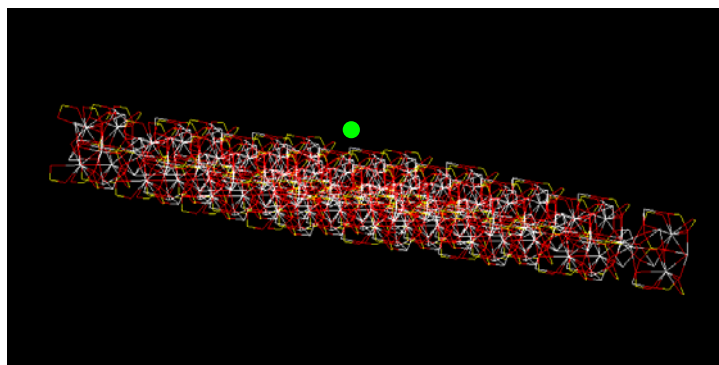


Fig. 3 – Positioning the reference atom in the vicinity of the crystalline face.

## RESULTS AND DISCUSSION

The script was started and, upon completion, a result file was obtained, and it contains the electric charges (C) and distances between successive atom pairs (Å) (reference atom-net atom):

atom-charge(1, 1) = 2

named-selection-value(1) = 20.86089

atom-charge(2, 1) = -1

named-selection-value(1) = 22.99303 .....

Figure 4 presents an example for the electric potential calculation as a summation of the influence of 5 randomly chosen  $\text{Ca}^{2+}$  ions from the crystalline net, in a reference point R situated in a plan parallel with the crystal's face at a distance of 2.1 Å. This influence,  $V_R$ , corresponds to the sum:

$$V_R = \sum_{i=1}^5 \frac{q_i}{d_i} = +2e\left(\frac{1}{d_1} + \frac{1}{d_2} + \dots + \frac{1}{d_5}\right) = 2 \cdot 1.6 \cdot 10^{-19} \text{ C} \left(\frac{1}{13.43\text{Å}} + \dots + \frac{1}{12.49\text{Å}}\right) = 9.59 \cdot 10^{-9} \text{ V} \quad (3)$$

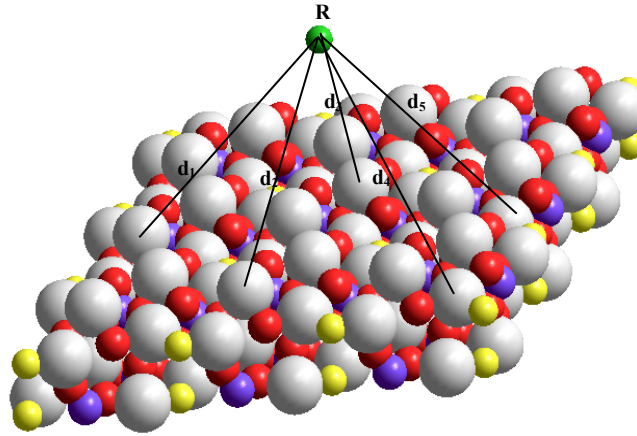


Fig. 4 – Example for the electric potential calculation for 5 randomly chosen atoms.

In a similar way, one can calculate the influence of all ions on reference point R.

Results for processing the influence of all 378 constituent ions of the crystalline face leads to a mathematical number that represent the electrostatic potential in the reference point R, at a considered distance  $d$  (Table 1).

Six reference points were chosen in the central zone of the crystalline face 110, electrostatic potentials were calculated and the arithmetic mean was obtained. This mean value was compared with a similar mean value obtained for the crystalline face 011, in order to determine the preferential direction for the crystal growth.

Table 1

Calculation of the electric potential  $V_R$  given by all constituent ions of the crystal's face;  $q$ -classical ionic charge (u.e.);  $d$ -distance to the reference point (Å);  $q/d$ -individual contribution to the collective potential  $V_R$  (volts)

No.	ionic charge $q$ (C)	distance $d$ to the reference point (Å)	electrostatic potential $q/d$ (V)
1	2	20.86089	0.095873
2	-1	22.99303	-0.04349
3	-2	20.69601	-0.09664
4	2	12.99226	0.153938
.....	.....	.....	.....
377	1	16.15221	0.061911
378	2	15.26604	0.131009
			summative potential = 5.19

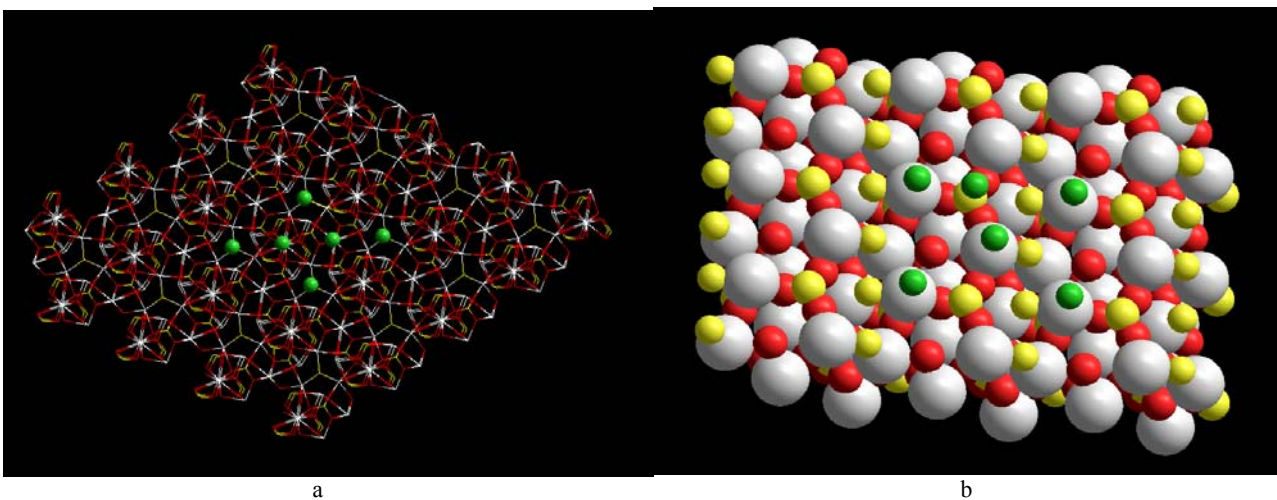


Fig. 5 – Reference points in the reference plan parallel with crystalline faces 110 (a) and 011 (b).

The results of electrostatic potential calculations at 2.1 Å distance from the crystal faces 110 and 011 are as follows:

110 face: 5,70; 5,51; 5,19; 4,97; 5,03; 4,98.  
Arithmetical mean: 5,23 V,  
011 face: 3,38; 3,37; 4,02; 3,71; 3,72; 3,74.  
Arithmetical mean: 3,65 V.

Modifying the distance between the reference plan (with reference points) and the crystalline face can reveal information concerning the variation of this potential at different distances near the crystal. Thus, for the crystalline face 110 the mean values for the electrostatic potential at the distances of 1.05, 2.1 and 4.2 Å are as follows in Table 2.

Results show a decrease of the electrostatic potential, and consequently a decrease of electric field influence with increase in distance from the crystalline face.

For an estimation on electrostatic potential variations over the whole crystalline face, its values were calculated along a space direction parallel with the ox cartesian axe, at equidistant reference points. Again, calculations were performed for three situations: at 1.05, 2.1 and 4.2 Å distances to the crystal.

Using the calculation protocol described above, electrostatic potential in each of the eight equidistant reference points was obtained, for each of the three considered reference planes, situated respectively at 1.05, 2.1 and 4.2 Å from the crystal's 110 face. The variation of the potential along the considered direction can be shown by nonlinear interpolation (Fig. 7). The interpolation curves were verified by calculating potential values corresponding to other (not shown) reference points.

Table 2

Electrostatic potential near the crystal face, at different distances	
distance to reference plan (Å)	mean value of the electric potential (V)
1.05	5.64
2.1	5.23
4.2	4.65

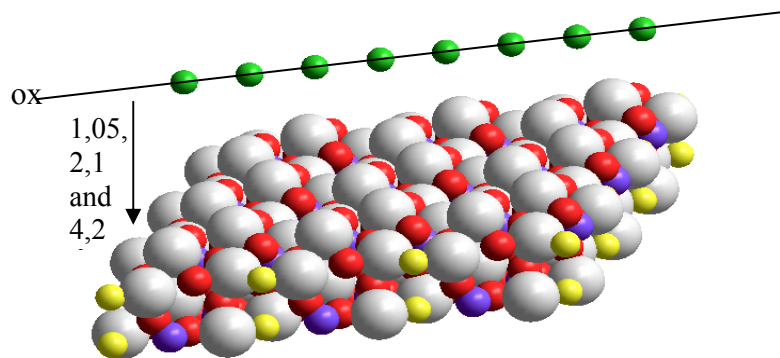


Fig. 6 – Reference points along the parallel ox direction.

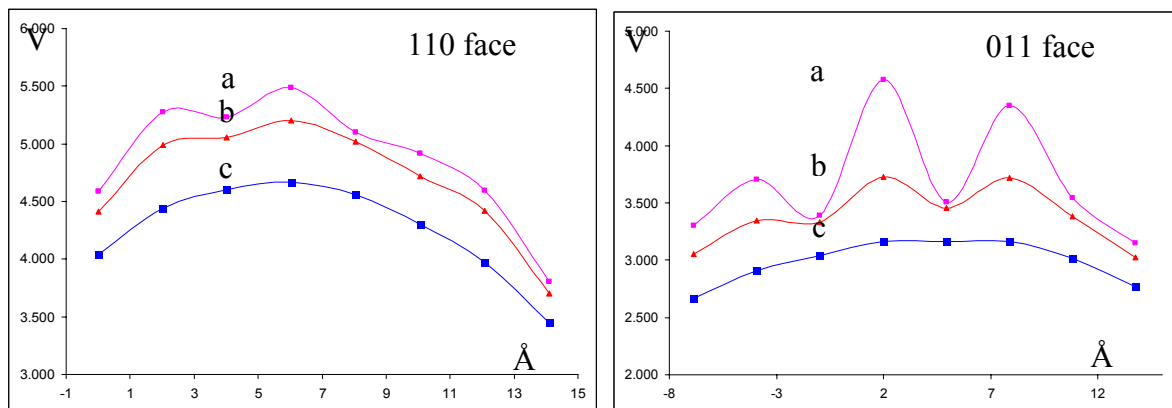


Fig. 7 – Electrostatic potential points along a parallel to the ox direction near the 110 face and respectively 011 face at: a) 1,05 Å; b) 2,1 Å; c) 4,2 Å.

## CONCLUSIONS

The calculations presented here show that the difference in electrostatic potential on the two different faces responsible for crystal growth is about 30% in the favor of the 110 face which is perpendicular to the growing crystal's direction, in agreement with the general principle that the face with the highest electric potential is the one to determine the direction of growth.

Electric potential on both surfaces is a positive value, so it can be assumed that first anions like  $\text{PO}_4^{3-}$  and  $\text{F}^-$  will be adsorbed from environment, followed by cations like  $\text{Ca}^{2+}$  in a consecutively way until a new layer is formed. Tensioactive substances (anionites, Lewis bases) will be adsorbed on to the most positive surface, the one with the rapid growth – reducing the growing speed of the crystal – resulting in small size, less elongated crystals. Collagen and other in vivo proteins from natural bone environment probably behave in a similar way. Relatively high content in  $\text{Mg}^{2+}$  ions in comparison to  $\text{Ca}^{2+}$  (about 1:30 in ions number) which are present in the bone hydroxyapatite could correspond to the competition between the two ions in the crystallization process; obviously, a too high percent of  $\text{Mg}^{2+}$  in hydroxyapatite will lead to the weakening, modification or even destruction of the crystalline net. The natural process for this

competition in bone environment is a complex one. If the smaller radius of  $\text{Mg}^{2+}$  ion could be a kinetically favorable factor for its adsorption in the crystalline net, and its diffusion through the surface protein layer, the more difficult dehydration process compared to  $\text{Ca}^{2+}$ , would be against a favorable absorption of  $\text{Mg}^{2+}$  in the crystal.

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