

## Effect of Magnesium on Biomineralization of Bone

Zhaolong Huang<sup>1,2, a\*</sup>, Shaoping Feng<sup>1,2,b</sup>, Ying He<sup>1,2,c</sup>, Mengshu Lai<sup>3</sup>,  
Yan Jiang<sup>1,2</sup>, Ruiming Xiao<sup>1,2</sup>, Lida Sun<sup>1,2</sup>

<sup>1</sup>School of Science, Honghe University, Mengzi, Yunnan 661100, P. R. China

<sup>2</sup>Key Laboratory of Natural Pharmaceutical & Chemical Biology of Yunnan Province, Yunnan 661100, P. R. China

<sup>3</sup>The State Administration for Entry-Exit Inspection and Quarantine of Hekou, Hekou 661300, China

<sup>a</sup>zhuang0132@126.com, <sup>b</sup>shaopingfeng@126.com, <sup>c</sup>heyeng@126.com,

\*corresponding author: zhuang0132@126.com

**Keywords:** biomineralization, chicken bones, calcium content, phosphorus content

**Abstract.** The phosphorus content, molar ratio of calcium and phosphorus and the effects of magnesium on calcium content in chicken bones were studied, comparing with different three groups which was fed with magnesium salt. The result showed that calcium content and phosphorus content of experimental groups have raised compared with those of normal group and the molar ratio of calcium and phosphorus of experimental groups decreased with the extension of feeding time. In addition, the molar ratio of calcium and phosphorus of experimental group, including liquid group and solid group which was fed with magnesium salt, decreased slower than that of normal group. A process that mixture of containing-phosphate and consisted of non-phosphate calcium salt changed into hydroxyapatite, calcium phosphate and calcium hydrophosphate, had happened.

### Introduction

Biomineralization is an important process in the formation of hard tissues in human and other vertebrates. Not only amounts of calcium, phosphorus and collagen are required, but also regulatory protein factors, enzymes and other biological activation component have play an important role on regulating and inducting the process of biomineralization of bone[1,2]. About 74% of dry bone is inorganic constituent (principal components, hydroxyapatite, HA) and rest of dry bone is organic matter (principal components, collagen). It is known that there is a close relationship between magnesium ion and many proteins, nucleic acid, enzyme structure, metabolism, but the particular effects which magnesium ion played on bone tissue growth and natural environment biological mineralization of collagen in bone wasn't reported frequently. In literature review, C.M. Serre pointed out that magnesium ion could slow down the degradation of calcium phosphate and high concentration of magnesium ion be poisonous to bone cells [3]. L.P. Zhao reported that magnesium avail fracture healing[4,5]. It was found that magnesium has an effect on the process of collagen self-assembly mineralization and the process was slow down on the condition that the magnesium existed in bone in our study[6]. In this paper, the influences of magnesium ion on calcium, phosphorus levels and inorganic components in chicken bones were studied and some rules were found during chicken growth.

### Experiment

**Materials and Methods.** Live chicken 27 bought from the local markets, which had grown for one month from hatch, and they were similar each other in age and weight. Other inorganic reagent were analytical reagent.

**Experiment.** 27 chicken was grouped into 3 groups, the first group was normal group (marked NG), the second group was fed solid MgSO<sub>4</sub>(marked SG) and the third group was fed aqueous solution of MgSO<sub>4</sub> (marked LG). SG and LG ate 0.43 gram of magnesium every day. Nine chicken

which every three chicken come from the same group (NG, SG and LG ) was killed and removed out the bones to test elements every month.

Elements analysis provided by Honghe agricultural comprehensive testing center. Test standard: GB/T5009.87-2003, GB/T5009.91-2003, GB/T5009.92-2003, GB/T5009.90-2003.

## Result and discussion

**Variation of calcium content with magnesium in chicken bones.** We analyzed the calcium content of chicken bones at 2 months, 3 months and 4 months respectively, and the results were shown in Table 1.

**Table 1 Calcium content in the bone during growth period**

Growth period (month)	Ca content(%)		
	liquid	solid	normal
2	6.91	6.89	6.62
3	7.78	6.90	6.75
4	7.69	6.98	7.11

As it can be seen from Table 1, the content of bone calcium of the liquid group(LG) fed with the magnesium salt solution is higher than the normal group(NG), while the calcium content of the solid magnesium group(SG) and the normal group(NG) isn't relatively obvious. It showed that the content of bone calcium has raised with the increasing of the content of magnesium, this is being that it is the magnesium salt solution that the chicken can be easy to absorb, while the solid magnesium salt is not obvious, as the overwhelming reason being that absorption of solid magnesium salt might be difficult to the chicken.

A possible reason why the calcium content could be increase when chicken have ate additional magnesium salt is that there was an antagonism between serum calcium and serum magnesium(both of them could be form complex compound). The antagonism could accelerate free calcium ion into the bone cell to form HA when the concentration of serum magnesium increased. Another reason was that activity of some coenzyme increased by magnesium, accelerating the transference of calcium.

Calcium content in SG was high slightly compared with NG, but it was lower than that in NG when chicken were fed to 4 month, as the reason was not known. The calcium content in LG was always higher compared with SG and the reason explained above.

It should be indicated that bone mineral density, bone strength will raise with the increasing of calcium content of bone, this is being that magnesium ion could accelerate the increase of bone mineral density.

**Variation of phosphorus content with magnesium in chicken bones.** The effect of phosphorus content in chicken bones with different magnesium salt was showed in Table 2.

**Table 2 Phosphorus content in the bone during growth period**

Growth period (month)	P content(%)		
	liquid	solid	normal
2	2.17	2.58	2.04
3	3.62	4.18	4.20
4	4.82	3.63	4.63

Experiments in table 2 showed that the phosphorus content of three groups in the bone raised with the increase of feeding time. In addition, the phosphorus content of the LG was slightly higher than the NG in February and April, the phosphorus content of the SG was significantly more than that the NG at the beginning of feeding time, but was unnatural in the coming time, especially in April, as the

reason may be related to poor solid magnesium absorption efficiency. The change of phosphorus content with time for LG showed that feeding magnesium salt was helpful to increase phosphorus content in chicken bones.

**Variation of molar ratio of calcium to phosphorus with magnesium in chicken bones.** The effect of molar ratio of Ca and P in chicken bones with magnesium salt was showed in Table 3.

**Table 3 Variation of molar ratio of Ca to P during feeding**

Growth period (month)	n(Ca)/n(P)		
	liquid	solid	normal
2	2.47	2.07	2.52
3	1.66	1.28	1.25
4	1.24	1.49	1.19

As seen from table 4, the molar ratio of Ca to P (MRCP) in the bone decreased with the feeding time in three groups and the decline for LG was slower than other both groups. In the same growth period, the MRCP of two groups of LG and SG were higher than the control group(NG) except the second month, the group of LG was close to the NG and the SG was lower than the NG.

It was known that normal inorganic composition in the bones is hydroxyapatite ( $\text{Ca}_{10}(\text{OH})_2(\text{PO}_4)_6$ , marked HA), molar ratio n(Ca)/n(P) is equal to 1.67 in HA. Analysis of growth rule in chicken bones, it was found that MRCP in three groups exceeded 1.67 when feeding for 2 months in three groups, regardless of feeding or not. Generally believed that the new bone may partially exist ( $\text{CaHPO}_4$ , the MRCP was 1) in the form of calcium hydrogen phosphate, and then gradually transformed into hydroxyapatite (HA, the MRCP was 1.67), but the old chicken bones calcium phosphorus ratio is higher than that of 1.67, the possibility reason being that calcium compounds of chicken may also exist in other[7].

From the beginning of the third month, MRCP in three groups was below normal value 1.67 and the LG was the maximum value near 1.67. MRCP of LG was reduced to 1.24 which was still higher than the NG (1.19) at the fourth month, while the SG was 1.49, as might be related with feeding solid salt and absorption abnormally.

Analysis of the change of MRCP during feeding from 2 to 4 month, variation of MRCP for LG was 1.23 and for NG was 1.33, indicating that bone mineral composition may tend to be more the ACH of the smaller ratio. The constituent in bone is HA when MRCP was 1.67:1, while is ACH when MRCP was 1:1, as is indicating that more HA existed in chicken bones with feeding magnesium salt. Therefore, we conclude that magnesium element could promote the stability of chicken bones and decrease the ratio of HA decomposition.

**Variation of magnesium content in chicken bones.** The variation of magnesium content in the bone was showed in Table 4.

**Table 4 Magnesium content in the bone during growth period**

Growth period (month)	Mg content(%)		
	liquid	solid	normal
2	0.94	0.96	0.89
3	1.27	1.14	0.89
4	1.62	1.50	1.50

As seen from Table 4, magnesium content in the bone for LG and SG was more than NG, and LG was high compared with the SG. It is generally believed that magnesium ion could replace calcium ion at the same position in the bone and cause the change of bone property, but the calcium content in

bone also increased, this is being that the increase of magnesium can not enough influences chicken growth of bone calcium content with the the growth of chicken. The reason of the increase of calcium content in chicken bones might be that magnesium can accelerate the activity of some coenzyme in bone and promote the calcium ions migrate into the interior of bone cells, except replacing of calcium ion. It is known that the more of serum magnesium concentration, the higher of carboxyl that was bonded at serum, which weakens the complex of calcium and serum carboxyl, accelerating the migration of calcium ion to bone cells and increasing the content of bone calcium.

## Summary

Magnesium played a role on increasing calcium and phosphorus content in chicken bones. Soluble magnesium salt is better than solid salt to rise calcium and phosphorus content in the bone and the effect on the calcium content is more than phosphorus content. MRCP of chicken bones in three groups decreased with the extension of feeding time and the decline for LG was slower than other both groups. It was speculated that some change process had happened, as  $\text{CaCO}_3$  change into HA and HA+ACH, which may be related to the process of growth and faded about the chicken. Although magnesium content in the bone increased with the extension of feeding time after feeding magnesium salt, the effects of magnesium on chicken bones composition were mainly derived from the factors on the role of coenzyme and other biochemical factors.

## Acknowledgements

This work was financially supported by the Science and Technology Support Program of China (61361002 ) and by Yunnan Province Science and technology Projects (2013FZ123).

## References

- [1] M.A. Dupree, S.R. Pollack, E.M. Levine, et al: *Biophys J*, 2006, 91(8): 3097
- [2] S. Gallea, F. Lallemand, A. Atfi, et al.: *Bone*, 2001, 28(5):491.
- [3] C.M. Serre, M. Papillard, P. Chavassieux , et al: *International Journal of Inorganic Materials*. 2000,(2): 455-461.
- [4] L.P. Zhao, SX Fang. *Journal of Traditional Chinese Veterinary Medicine*, 2002, (5): 17-19.
- [5] M.J. Xun, X.K. Wang, A. Wang. *Journal of Northeast Agricultural University*, 1999, 30(4): 355-358.
- [6] Z.L. Huang, Y. He, Q. Cai. et al: *Journal of Honghe University*, 2005, 3(3): 1-4
- [7] H.Q. Gu, G.F. Xu. *Biomedical Materials*. Tianjin Science & Technology Translation & Publishing Corp, Tianjin, 1993, pp. 54-59.