

# Fourier Transform Infrared Spectroscopic Analysis of Bone of Chicken *Gallus gallus domesticus*

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**ABSTRACT:** The paper reports FTIR data of powdered bone of chicken *Gallus gallus domesticus*. Fresh samples of bone are separated carefully from the flesh, cleaned thoroughly and sun dried to remove all the water and powdered for the FTIR analysis. FTIR spectrum reveals characteristic bands concerned with protein, lipids and carbohydrates. Further, the major inorganic constituent, calcium phosphate is evident from the characteristic bands of functional groups.

**KEYWORDS:** FTIR spectroscopy; Chicken; Bone; Calcium Phosphate.

## I. INTRODUCTION

FTIR is a technique used to obtain an infrared spectrum of a solid, liquid or gas. It is a powerful analytical instrument for detecting functional groups and characterizing covalent bonding data. Fourier transform infrared spectroscopy (FTIR) identifies chemical bonds in a molecule by producing an infrared absorption. A distinctive molecular fingerprint is used to screen and scan samples for many different components.

Bone is zone of the most sophisticated biological structures developed by nature [1]. The major constituent of bone is calcium phosphate mineral which is similar in composition and structure to minerals within the apatite group, which form naturally in the Earth's crust [2]. Louisfert and Pobbequin [3] studied a variety of natural and synthetic samples of calcium carbonate, both individually and in mixture by infrared spectroscopy.

Vijaya Ushasree, et al [4] reported FTIR data on normal and diseased human blood and its constituents at different physiological conditions. They showed FTIR characterization of blood groups. Further they found significant difference in FTIR data and concluded that IR data can be used comfortably for the disease analysis, if clinically correlated.

Gangadhar et al [5] reported, based on FTIR spectroscopy, that the constituents of ovine scapular cartilage are mainly collagen and proteoglycans; Carbonate ions and phosphate ions are in very small quantity.

Nazima Siddiqui and Adeel Ahmad [6] presented IR spectroscopic data of *Ten* edible and *Fifteen* medicinal oils of plant origin for IR analysis. The FTIR spectra of edible and medicinal oils showed a series of bands with different intensities and revealed the composition of fatty acids and degree of saturation of the selected oils. The study suggested that IR spectroscopy could be considered as a vital technique for identification, analysis, determination of degree of saturation of fatty acids and detection of adulteration of oils of plant origin.

Syed Ismail Ahmed, et al [7] analysed FTIR spectra in the mid IR region for normal and albumin treated urine. It was observed that peaks at  $1641\text{ cm}^{-1}$  and  $1450\text{ cm}^{-1}$  in IR spectra were the most specific peaks for urea and albumin, respectively. They concluded that FTIR method of detecting proteinuria is quick and cheaper.

Masood Mahmoodi, et al [8] presented IR spectroscopic data on sheep skeletal muscle. The study revealed the presence of contractile proteins, phospholipids, triglycerides and cholesterol along with phosphates in the skeletal muscle.

Posner et al [9] carried out infrared studies on synthetic hydroxyl apatite  $\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$ , containing low calcium and the presence of fairly strong hydrogen bonds in proportion to the missing number of calcium ions.

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Srinivasa Rao and Srinivasa Manja [10] studied infrared spectrum of human bone powder and ash with a view to understand the nature of hydrogen bonding in these biological apatite. The infrared spectrum revealed bands of characteristic of CO<sub>3</sub> ion and this observation suggested that bone material consists of carbonate containing apatite. The average bond lengths in bone powder and bone ash were estimated to be 2.76 Å and 2.80 Å respectively.

A search of literature reveals that no FTIR spectroscopic studies are available on the bone of chicken *Gallus gallus domesticus*. In view of this, an attempt is made to study chicken bone spectroscopically using the technique of FTIR spectroscopy.

## II. MATERIALS AND METHODS

The bones samples of *Gallus gallus domesticus* (chicken) were carefully separated from the flesh, cleaned with water thoroughly and sun dried so as to remove free water completely. The dried bone samples were powdered using pestle and mortar for the use of FTIR analysis.

Infrared spectrum of the bone powder was recorded in FTIR spectrometer (Shimadzu FTIR - 8400S) in the range of 4000 cm<sup>-1</sup> to 400 cm<sup>-1</sup>. The powdered sample of bone was mixed with potassium bromide (KBr) in the ratio of 4:1 and pressed in a stainless steel die to produce thin KBr wafer, containing a relatively high concentration of the sample in IR transparent KBr matrix. The sample underwent ten scans.

## III. RESULTS AND DISCUSSION

Fig. 1. shows FTIR spectrum of bone of chicken *Gallus gallus domesticus* recorded in the range of 4000 cm<sup>-1</sup> to 400 cm<sup>-1</sup>. It reveals a series of bands with different intensities. The data obtained from FTIR spectrum is tabulated in Table 1 for analysis.

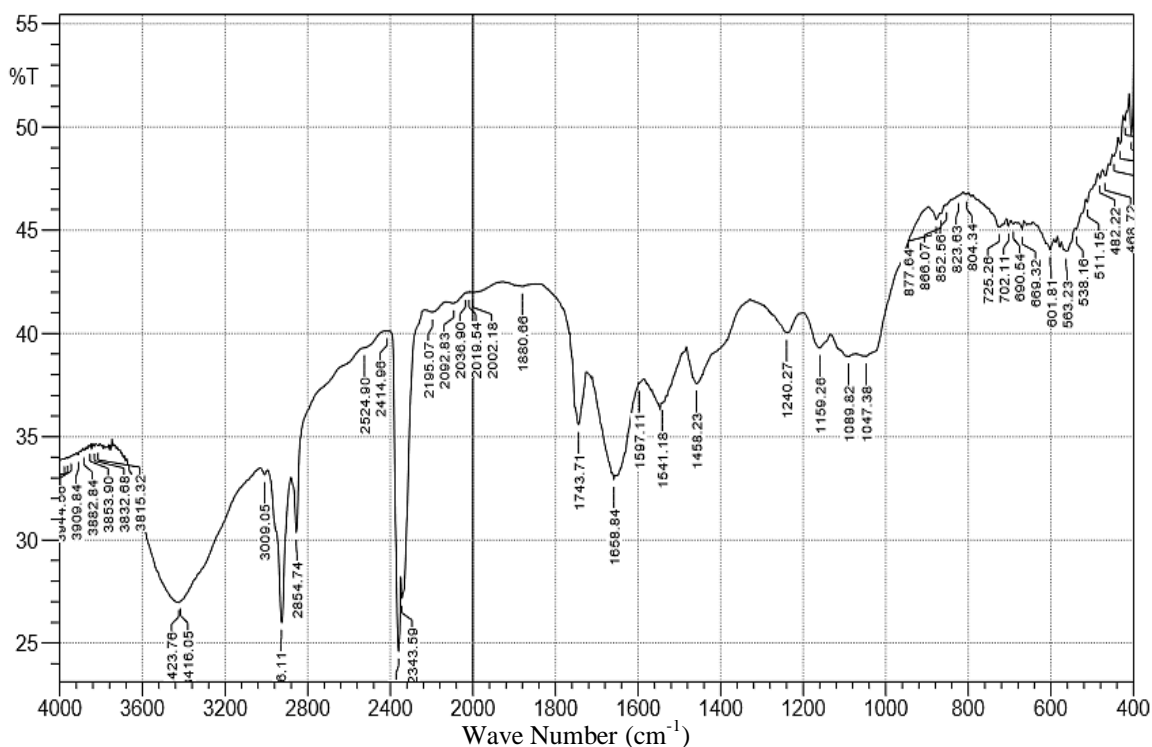


Fig. 1. FTIR spectrum of bone of chicken *Gallus gallus domesticus*

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The IR spectrum serves as a 'finger print' of biological macromolecules present in the sample. It is to be noted that the intensities (% transmittance or % absorbance) of IR spectral bands provide quantitative information, while their absorption positions (in wave number) reveal qualitative information on the nature of chemical bonds, their structure and their molecular environment.

For the IR analysis of a sample, IR spectrum is divided into three broad regions. Region I (4000 - 3000  $\text{cm}^{-1}$ ) reveals the nature of hydrogen bonding. Region II (3000 - 1500  $\text{cm}^{-1}$ ) is pertaining to functional groups present. Region III (1500 - 400  $\text{cm}^{-1}$ ) gives information about most of the bio - minerals and their combinations, phases and substitutions.

Table 1 – FTIR data on the bone of chicken *Gallus gallus domesticus*

Wave Number ( $\text{cm}^{-1}$ )	T (%)	Characteristic functional groups
3424 3416	26.97 20.30	NH <sub>2</sub> ; Carbonyl C=O stretching band; O-H stretching mode
3009	33.16	CH stretching vibration of the cis double bond (=CH) within unsaturated fatty acyl ester.
2926	25.99	-C-H symmetric stretching of -CH <sub>3</sub>
2855	30.36	-C-H symmetric stretching of -CH <sub>2</sub>
2343	27.15	Atmospheric absorption; CO <sub>2</sub>
1743	35.57	C=O stretching vibrations of ester functional groups in phospholipids;
1658	32.92	Amide I; $\alpha$ - helical structure (C=O)
1541	36.63	C-O-anti symmetric stretch
1458	37.58	CH <sub>2</sub> bend; P-O- anti symmetric stretch
1240	40.04	Amide III band components of proteins (C-N)
1159	39.31	C-C vibrations
1089	38.89	P=O symmetric stretching vibrations of PO <sub>2</sub>
1047	38.90	Carbohydrate bands suggests C=O absorption of glycoproteins
877	45.53	C-O out of plane bending
602	44.05	Phosphorus (P <sub>4</sub> )
563	43.97	P-O- anti symmetric bend
468	47.63	P-O- symmetric bend

In the present study, major absorption band arise from the functional groups such as NH, C=O, C-H and P=O concerned with proteins, lipids and nucleic acids. The region between 4000 – 3000  $\text{cm}^{-1}$  is dominated by broad spectral characteristics bands (3424; 3416  $\text{cm}^{-1}$ ) arise from NH<sub>2</sub>, Carbonyl C=O; O-H stretching modes.

The region between 3100  $\text{cm}^{-1}$  and 2800  $\text{cm}^{-1}$  reveals C-H stretching vibration of CH<sub>3</sub> and CH<sub>2</sub> functional groups and hence dominated by fatty acids and also by some amino acid side chain vibrations. The band about 1458  $\text{cm}^{-1}$  in the region between 1470  $\text{cm}^{-1}$  and 1350  $\text{cm}^{-1}$  is concerned with various deformation modes of the functional groups.

Further, the bands at 1744  $\text{cm}^{-1}$ , 1658  $\text{cm}^{-1}$ , 1597  $\text{cm}^{-1}$  and 1541  $\text{cm}^{-1}$  in the region between 1800  $\text{cm}^{-1}$  and 1500  $\text{cm}^{-1}$  are dominated by conformation sensitive Amide – I and Amide – II bands, which are very sensitive bands as far as biological complex systems are considered. As is known, IR spectroscopy is an averaging technique and as such Amide – I and Amide – II bands cannot provide structure information of a particular protein, but can suggest the dominant conformation  $\alpha$  or  $\beta$  or both.

In the mineralized tissues such as bone, integuments, shells etc, the mineral present in them can be calcium carbonate or calcium phosphate or both. In the present study, FTIR spectrum presents bands at 877  $\text{cm}^{-1}$  (C-O out of plane bending); 1541  $\text{cm}^{-1}$  (C-O - anti symmetric stretch) are characteristic of carbonate ion (CO<sub>3</sub><sup>2-</sup>).

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The band  $712\text{ cm}^{-1}$  characteristic of calcium carbonate ( $\text{CaCO}_3$ ) is absent. Hence, it can be concluded that free calcium carbonate is not present in chicken bone of the present study.

The bands  $468\text{ cm}^{-1}$  (P-O - symmetric bend),  $563\text{ cm}^{-1}$  (P-O- anti symmetric bend),  $1089\text{ cm}^{-1}$  (P-O-symmetric stretch),  $1458\text{ cm}^{-1}$  (P-O- anti symmetric stretch) are pertaining to phosphate ion ( $\text{PO}_4^{3-}$ ). It is important to note that there is no split of  $603\text{ cm}^{-1}$ , which suggests that calcium phosphate present in chicken bone is mostly amorphous and less crystalline.

### IV. CONCLUSION

The present study reveals the fact that, the mineral present in chicken bone is hydroxyl calcium phosphate. Further, calcium phosphate present in bone is of both crystalline and amorphous in nature. The same has been studied for other species like fishes.

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