Serum alkaline phosphatase level in acromegaly patients and an association with its level in saliva

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Serum Alkaline Phosphatase Levels in Acromegaly Patients and an Association With Salivary Levels

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Abstract

Objective: The study aimed to determine the levels of bone turnover biomarker (ALP) in Iraqi acromegaly patients in addition to finding the relationship between these levels in serum and saliva.

Methods: In this cross-sectional study, 20 healthy controls (10 female and 10 male) and 40 patients with acromegaly (22 female and 18 male) who were visiting the National Centre for Diabetes and Endocrinology/Al-Mustansiriya University/Baghdad were included. Every patient was prescribed an octreotide somatostatin analog medication after being diagnosed by a hospital doctor. For both groups, the levels of alkaline phosphatase (ALP) in serum and saliva were measured.

Results: The acromegaly group had higher mean values of serum and salivary ALP than the control group (p = 0.01, p < 0.001, respectively).

Conclusion: High significant correlation has been found between the concentrations of serum and salivary ALP in acromegaly patients while the correlation coefficient between the last concentrations in control subjects was with non-significant association.

Keywords: Acromegaly, Bone formation marker, ALP, Salivary alkaline phosphatase, Alkaline phosphatase

Acromegaly is a rare disease by increased growth hormone (GH) and insulin-like growth factor 1 (IGF-1). In the vast majority of instances, pituitary adenomas are the reason. Acromegaly patients experience several comorbidities and metabolic issues as a result of elevated levels of GH and IGF1 [1,2]. Because the disease is so subtle, the clinical diagnosis, which is based on GH rise symptoms, is frequently made later than necessary [3].

Through both direct and indirect effects via IGF-, GH is crucial for the growth, differentiation, and repair of bone and cartilage production [4]. An important function of the GH and (IGF-I) is to monitor early maturation and the health of the skeleton. Growth hormone (GH) is essential for the development of top bone mass, which is the primary indicator of the risk of osteoporotic fractures. Additionally, through the management of bone turnover, GH plays a crucial part in maintaining the architecture of the skeleton throughout adulthood [5]. It is well known that the activity of the bone tissue's alkaline phosphatase indicates bone development [6]. Measured as protein content or as enzyme activity, total alkaline phosphatase (ALP) and bone alkaline phosphatase (BALP) are commonly utilized as indicators of bone formation, especially in patients with primary and secondary bone disorders and disruptions in calcium–phosphorus equilibrium.

ALP and BALP are useful indicators of primary bone tumors and bone metastases. They also aid in...
the early diagnosis of malignancies connected to the bones and the monitoring of the therapeutic effects of oncologic therapy [7,8]. The enzyme alkaline phosphatase (ALP) is widely distributed in several human tissues, namely in the intestines, kidneys, bones, liver, and teeth [9]. It is known as alkaline phosphatase as it is responsible for removing the phosphate group from proteins and other biomolecules. It needs a basic PH of 10 to function at its best. Its role is essential to the wellness of the body. This enzyme exists in several isoforms, the most significant ones being intestinal, placental, liver, and bone ALP [10]. Compared to children, adults have lower amounts of ALP because children's developing bones create larger levels of ALP. There are instances where levels might reach 500 IU/L during growth spurts [11]. Changes in blood alkaline phosphatase levels may be suggestive of particular diseases. The primary use of ALP measurement is to identify liver or bone problems [12].

Saliva is a biological fluid (biofluid) that may be used for laboratory or clinical diagnosis, prognosis, and monitoring and treatment of patients with systemic and oral disorders. It is a clinically informative bio-fluid [13]. The ease and noninvasiveness of saliva sample collection significantly reduces the discomfort associated with blood collection and the privacy concerns connected with urine collection, making it a valuable diagnostic tool [14]. For both the patient and the operator, oral fluid collection is safe. These features allow for the monitoring of several biomarkers in young children, adults, the elderly, and non-collaborative patients, as well as in many situations when it is not feasible to collect blood or urine samples [15].

2. Materials & methods

Twenty young, healthy volunteers (ten females and ten male, ages 29 to 54) and forty acromegaly patients (sixteen females and eighteen males, ages 28 to 63) were recruited, all patients were chosen from among those who visited the Mustansiriya University National Center for Diabetes and Endocrinology and were given an acromegaly diagnosis by an endocrinology specialist.

The diagnosis was made using the standard clinical presentation and laboratory parameters, such as high insulin-like growth factor and GH levels that did not reduce following an oral glucose load. None of the chosen patients had liver problems. Following an overnight fast, blood samples were taken from each individual between 8.0 and 11.0 a.m. The samples of blood were taken and centrifuged for 10 min at 3000 r/m. When it was time for analysis, the leftover serum was stored at -20° C.

Whole saliva that was at rest was collected between 8.0 and 11.0 a.m. Patients were instructed to refrain from eating, drinking, or using any oral hygiene products. They were also directed to rinse their mouths with water, produce saliva, and spit into a large test tube. After the saliva was collected, it was centrifuged for 10 min at 3000 r/m. Before being subjected to laboratory investigations, the resultant supernatant was kept at -20° C. Sample analyses were conducted at the Mustansiriya University, National Center for Diabetes and Endocrinology. For alkaline phosphatase quantitative determination, Serum and whole saliva were assessed colorimetrically by a spectrophotometer (liquicolor kit from human company Germany was used for this purpose).

- Working reagent was prepared by add one ml from substrate to bottle contained 4 ml buffer.
- The spectrophotometer setting adjusted as: Wavelength: Hg 405 nm (400–420 nm), Optical path: 1 cm.
- The temperature kept constant (±0.5 °C) during the duration of test.
- The procedure was done as: pipetted 20 μl of sample and then 1000 μl of working reagent, mixed then read the absorbance after 1, 2, 3 min. Then the mean of the reading was calculated.

2.1. Statistical analysis

DATA description, analysis and presentation were performed using two computer software programs, Statistical Package for Social Sciences (SPSS) version 16 and Microsoft Office Excel 2007. Mean, SD, and SE were used to represent numerical variables, while frequency was used to represent nominal variables. The mean numerical variable between the two groups was compared using the t-test. The correlation between numerical variables was assessed using Pearson’s correlation coefficient. Finally accepted level of significance for p-value was considered (0.05).

3. Results

The clinical parameter mean, standard deviation, and standard error are shown in (Table 1) for both healthy and acromegaly patients shown that individuals with acromegaly had higher mean ALP values than did healthy participants, and the p-value of (0.013) indicated a significant difference.

The results also show increasing the mean value of salivary ALP in acromegaly patients than healthy subjects and the p-value which was (<0.001) showed significant difference.
The correlation coefficient between the concentration of serum ALP and salivary ALP in acromegaly patients was \( r = 0.544 \) with highly significant association at \( p < 0.001 \) as shown in Fig. 1. While the correlation coefficient between the concentration of serum ALP and salivary ALP in control subjects was \( r = 0.394 \) with non-significant association at \( p > 0.05 \) as shown in Fig. 2.

### 4. Discussion

Serum alkalinephosphatase levels are measured as the traditional clinical indicator of bone metabolism [16], because it is easily accessible, affordable, and relatively easy to administer, it is the most often used measure of bone metabolism, after liver illness has been ruled out. Serum total ALP levels give an accurate indicator of the amount of osteoblast activity and new bone production [17].

This study investigated the (ALP) in acromegaly patients and the association between its level in serum and saliva. Serum ALP was found statistically significantly higher in patients with acromegaly compared to healthy controls. This finding agrees with Marazuela in 1993 and Mehmet in 2022 [18,19]. Also Many studies show that increase bone turnover, mainly formation, in Patient with acromegaly, Ezzat in 1993, Piovesan in 1994, Legovini in 1997, Bolanowski in 2002 and Morselli in 2006 [20–24].

In 2018, a study by Eiman discovered that, when compared to the control group, the group of patients with acromegaly had a highly significant spike in bone alkaline phosphatase (BALP) levels, which are indicators of bone formation [25]. Furthermore, Mehmet in 2022 found the relationship between bone biochemical markers and disease activity. Specifically, he found that patients with active acromegaly had greater levels of ALP compared to those who were receiving surgical or medicinal treatment for remission [19].

Unstimulated saliva were collected due to that during most parts of day and night the mouth will be protected by resting saliva alone and to avoid any changes in chemical composition of saliva by stimulation [26]. The subjects were instructed not brush their teeth or use any other kind of oral care for several hours before collecting a saliva sample to avoid tiny abrasions which lead to exudate the plasma in the mouth [27].

It was found that salivary ALP significantly increases in acromegaly patients than control subjects.

### Table 1. Mean, SD and SE of clinical parameters for healthy and acromegaly patients.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Acromegaly group</th>
<th>Control group</th>
<th>( P )</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>SD</td>
<td>SE</td>
</tr>
<tr>
<td>Serum ALP</td>
<td>156.82 U/L</td>
<td>44.16</td>
<td>6.98</td>
</tr>
<tr>
<td>Salivary ALP</td>
<td>11.61 U/L</td>
<td>2.70</td>
<td>0.43</td>
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Significant p-value (0.05); * significant difference. **highly significant difference.
This comes into agreement with Alfredo in 1997 [28]. In acromegaly patients, in the presence of GH excess, a high alveolar bone formation may occur [29,30], this due to the direct effect of GH on osteoblasts and via local production of IGF-I (autocrine/paracrine action) [31]. Desai in 2009 suggested that salivary ALP as a marker of alveolar bone remodelling [32].

Moreover, this study also showed significant correlation between salivary ALP and serum ALP in acromegaly patients. This comes in to agreement with Pellegrini in 2008 indicated that the plasma component leakage into saliva [33]. Several pathways may be involved in the clearing of chemicals from plasma into saliva:

(a) Ultrafiltration via intercellular nexus (gap junctions) between secretory unit cells. (b) Transudation of plasma chemical compounds into the oral cavity through the oral mucosa, crevicular fluid, or both. (c) Two methods of selective transport across cellular membranes include active transport via protein channels or passive diffusion of lipophilic substances (steroid hormones) [34].

5. Conclusion

In conclusion, the level of serum and salivary ALP were increased in acromegaly patients and high significant correlation has been found between salivary ALP and serum ALP.

References


