



The Effect of COVID-19 Infection on Human Blood Ghrelin Hormone: A Pilot Study

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Authors' contributions

This work was carried out in collaboration among all authors. Author NYH designed the study. Authors NYH, EOT and MMS conducted the technique. Authors HMAO, OFA and HAA focused on providing the materials of study as well as collecting and organizing data. Authors NYH and HAA provided data analyzes and interpretation. Authors NYH, WAA, HMAO and HAA worked together on writing the article. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JPRI/2021/v33i731199

Editor(s):

- (1) Dr. Ana Cláudia Coelho, University of Trás-os-Montes and Alto Douro, Portugal.
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Complete Peer review History: <http://www.sdiarticle4.com/review-history/65527>

Original Research Article

Received 20 December 2020

Accepted 26 February 2021

Published 03 March 2021

ABSTRACT

Objectives: The pandemic of coronavirus disease 2019 (COVID-19) has been one of the major health concerns for all the countries around the globe. This study was aimed to study the potential effect of COVID-19 virus on the level of blood ghrelin appetite hormone in order to determine the influence of this infection on the patient appetite.

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Methods: This is a cross-sectional study was conducted between July and the end of August 2020 in the western region of Saudi Arabia. A total of 50 confirmed positive patients with COVID-19 (positive group) and 30 control healthy subjects with negative blood samples (negative group) were collected to determine the level of ghrelin appetite hormone using the human ghrelin (GHRL) ELISA technique. A student's *t*-test was carried out to find out the statistical change between different study groups.

Results: The serum total ghrelin concentrations, on average, were 51.32 pg/mL on positive group and 50.37 pg/mL on negative group, respectively. The difference of ghrelin was statistically insignificant between the two groups ($P > 0.05$). Although the sample size of the study was small, the results showed high number of COVID-19 cases in male than female.

Conclusion: The current data shows that there are no significant changes in the level of serum ghrelin hormone in COVID-19 patients. Consequently, it might be possible that the ghrelin hormone showed potential changes in the saliva compared to the effect in the blood. Thus, a further analysis of the ghrelin hormone in the saliva of COVID-19 patients will be conducted in the near future.

Keywords: COVID19; ghrelin; respiratory symptoms; pandemic; appetite.

1. INTRODUCTION

The pandemic of coronavirus disease 2019 (COVID-19) has been one of the major health concerns for all the countries around the globe. During the past few months, most of the studies have focused on understanding this newly emerged coronavirus and its effect on the human health [1,2]. Recent reports have clarified the most common clinical symptoms of the infected individuals with COVID-19 [3-5]. One of these symptoms includes a loss of smell and food taste [4,5]. This may theoretically have a negative effect on the patient appetite. A multicenter European study has shown that some COVID-19 patients have experienced the loss of appetite along with other symptoms [5]. However, the mechanism of how the coronavirus has influenced the patient appetite is still unclear.

It is generally known that chronic smoking has adverse effects including loss of food taste, appetite, as well as weight [6,7]. A Previous pilot study has shown that the level of appetite hormone was significantly decreased in cigarette smoker [8]. The human appetite hormone, namely ghrelin, is normally secreted in the blood to improve the food intake and appetite through a complex neuroendocrine mechanism [9,10]. Moreover, the ghrelin hormone is an acylated peptide consisting of 28 amino acids and releasing from stomach wall. In addition, this hormone is secreted in the saliva and can control the food taste [10,11]. For example, it can decrease the sour taste and improve the sweet flavor [10-12]. Although the exact role of the ghrelin in saliva is still not fully understood, the salivary secretion of this hormone has shown an effect on the food taste and appetite [12].

The effect of COVID-19 virus on the level of blood ghrelin appetite hormone was investigated in this study, in order to determine the influence of this infection on the patient appetite.

2. MATERIALS AND METHODS

2.1 Samples Collection

A total of 30 volunteer and 50 COVID-19 positive subjects participated in this study. Venous blood samples were randomly collected from all 80 subjects (non-fasting) in plain tube under aseptic phlebotomy technique. Serum was collected and frozen at -20°C until the time of assay performing. The study was conducted in the western region of Saudi Arabia. An ethical approval was obtained and IRB number H-02-K-076-00520-298 has been granted by the Saudi Ministry of Health to work with patient samples in this study. All positive COVID-19 patients were confirmed by molecular PCR test.

2.2 Ghrelin Assay

Total ghrelin was measured in all samples using the human ghrelin (GHRL) 96 well-plate kit (BT, Shanghai, China). The assay was performed according to the manufacturer's procedure. Briefly, each sample was diluted 4x in the sample diluent and 100 μL was transferred into the corresponding wells. 50 μL from conjugate was added to each well except blank. The plate was covered and incubated for 60 minutes at 37°C . 50 μL from both substrate A and B was added to each well including blank. The plate was covered again and incubated for 20 minutes at 37°C . The reaction was stopped and the optical densities for standards and samples were determined at

450 nm using Bio Tek EL 340 microplate reader. The detection limit of ELISA technique has a wide range from 0.0 pg/ml to 100 ng/ml [13].

Graph was drawn using standards' absorbance values and their corresponding concentrations.

2.3 Statistical Analysis

Means and standard error of the mean (SEM) were calculated for each data set. Then an unpaired student's *t*-test was carried out to compare between different sets and *P* value <0.05 was considered statistically significant. GraphPad Prism software (San Diego, California, USA) was used to analyze the data in this study.

3. RESULTS

3.1 The Study Participants

As shown in Fig. 1, around 30 of non infected volunteers and 50 of COVID-19 positive patients have participated in this study. The number of male and female participants is equal (15 individuals) in the control group (Fig. 1a). Nevertheless, the number of COVID-19 cases is

clearly higher in male than female (Fig. 1b). Non-Saudi Participants who had COVID 19 were evidently higher than Saudis (Fig. 2b).

3.2 Blood Ghrelin

All data obtained were normally distributed. The mean \pm SEM values of total ghrelin in serum were 51.32 pg/mL and 50.37 pg/mL in control (healthy individuals) and COVID-19 positive (infected individuals) samples, respectively. The *t*-test comparison between the control and COVID-19 positive samples showed no statistical difference (*P* = 0.8127) in the level of blood ghrelin hormone between the two groups as shown in (Fig. 3).

4. DISCUSSION

The present data shows that COVID-19 virus did not significantly affect the level of ghrelin hormone. Therefore, the loss of appetite that was previously reported by COVID-19 patients yet required further explanation. In the light of these data, there was no direct association between the corona virus infection and the level of blood ghrelin hormone.

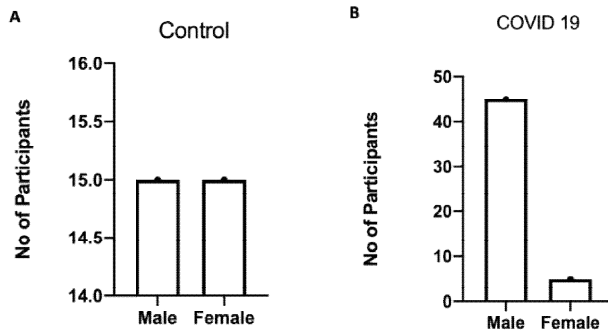


Fig. 1. Comparison between male and female participants in Control (A) and COVID-19 (B) groups, *n* = 30 and 50 for Control and COVID-19 respectively

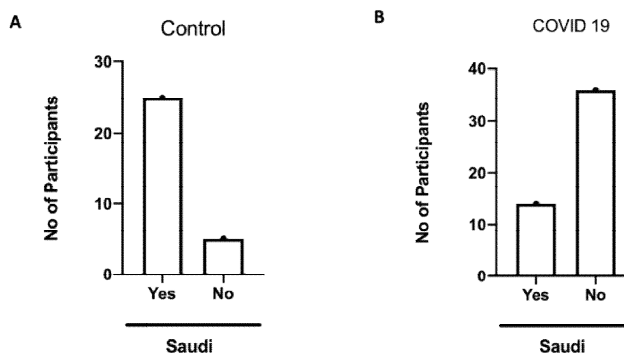


Fig. 2. Comparison between Saudi and non-Saudi participants in Control (A) and COVID 19 (B) groups

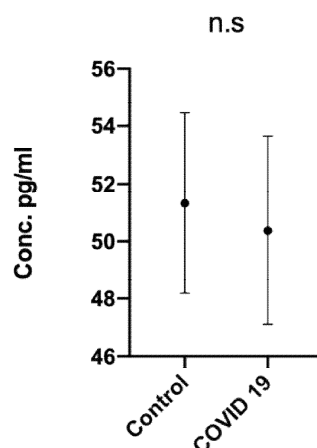


Fig. 3. Comparison between blood ghrelin levels in control and COVID-19 group showing no significant (n.s) difference between the two groups ($P > 0.05$ is considered insignificant as per *t*-test)

It is well known that the COVID-19 patient is not only associated with respiratory symptoms as it might be thought. Moreover, several digestive symptoms were well reported in latest reports [14]. Loss of appetite was documented as one of the digestive symptoms for many patients with confirmed COVID-19. A number of recent studies have reported the loss of appetite as one of the most frequent symptoms in COVID-19 patients [5,15,16]. A study by Lechien et al. has indicated that more than half of COVID-19 patients reported cases loss of appetite as one of their symptoms [5]. Similarly, other studies elucidated the presence of loss of appetite in more than 50% of COVID-19 along with gastrointestinal digestive symptoms [15,16]. In addition, it was reported as one of the most three common symptoms of COVID-19 infection [17]. Correspondingly, a multicenter European study has confirmed that the loss of appetite as a major clinical symptom for positive COVID-19 patients [5].

The appetite hormone ghrelin mainly produced and secreted in gastrointestinal pathway. It is an orexigenic hormone associated with nutrient sensing, food intake, and appetite [18]. The relationship between the serum ghrelin level and appetite was well documented [19]. An early investigation has determined a correlation between the level of ghrelin hormone and the hunger scores among healthy individuals [20]. Furthermore, previous study showed that the administration of ghrelin has enhanced the appetite and food intake in healthy subjects [21]. On the contrary, it is widely known that smoking can suppress the appetite. A previous

investigation evaluated the level of ghrelin in both blood and saliva samples among smokers who recently smoked one cigarette. Accordingly, the authors presented that the level of ghrelin hormone in the blood has no significant changes. However, the level of ghrelin in the saliva samples has significantly reduced [8]. Our current data shows that there are no significant changes in the level of serum ghrelin hormone in COVID-19 patients. Consequently, it might be possible that the ghrelin hormone showed potential changes in the saliva compared to the effect in the blood. Thus, a further analysis of the ghrelin hormone in the saliva of COVID-19 patients will be conducted in the near future.

This investigation is not without limitations as the sample size might be considered small. A larger sample size would definitely improve the generalizability of the study. Further, the analysis of saliva samples would add more strength to the current findings. Nevertheless, the levels of ghrelin hormone in the serum is used as marker of appetite.

5. CONCLUSION

The present investigation shows no changes in the levels of ghrelin hormone in the blood samples of COVID-19 patients. Consequently, the reported loss of appetite by COVID-19 patients warrants further investigations.

6. RECOMMENDATIONS

The reported digestive symptoms of COVID-19 patient is subject to revision and further investigations are needed.

CONSENT

As per international standard or university standard, patients' written consent has been collected and preserved by the authors.

ETHICAL APPROVAL

An ethical approval was obtained and IRB number H-02-K-076-00520-298 has been granted by the Saudi Ministry of Health to work with patient samples in this study.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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