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Research Article

Intra-day changes in the levels of biochemistry parameters

Abdullah Arpaci[1], Serdar Dogan[1], Bahar Unlu Gul[2], Berna Kus[3], Emre Dirican[4]

[1]Department of Medical Biochemistry, Mustafa Kemal University Faculty of Medicine, Hatay, Turkey
[2]Department of Medical Biochemistry, Kars Harakani Public Hospital, Kars, Turkey
[3]Department of Moleculer Biochemistry and Genetics, Mustafa Kemal University Faculty of Medicine, Hatay, Turkey
[4]Department of Biostatistics and Medical Informatics, Mustafa Kemal University Faculty of Medicine, Hatay, Turkey

Abstract

Objectives: People mostly live in the nonfasting state during a normal 24-h cycle. This study aims to compare the levels of 18 biochemical parameters during different hours of the day.

Methods: A total of 18 biochemical tests of patients who visited outpatient clinics only once between January 1, 2010, and December 31, 2019, were evaluated at the Hatay Mustafa Kemal University (HMKU) Central Laboratory by using hospital database information. The tests are albumin (Alb), aspartate aminotransaminase (AST), alanine aminotransaminase (ALT), alkaline phosphatase (ALP), amylase, blood urea nitrogen (BUN), calcium (Ca), total cholesterol (TC), creatine kinase (CK), creatinine (Cr), gamma-glutamyltransferase (GGT), glucose, high-density lipoprotein cholesterol (HDL-C), inorganic phosphorus (Pi), iron (Fe), total protein (TP), triglyceride (TG), and lipase. The blood samples of the patient were divided into eight groups according to their collection time as follows: (a) 07:00-07:59, (b) 08:00-08:59, (c) 09:00-09:59, (d) 10:00-10:59, (e) 11:00-11:59, (f) 12:00-13:59, (g) 14:00-14:59, and (h) 15:00-17:00.

Results: A statistically significant difference was found between the groups in terms of all parameters except amylase, GGT, and TP ($p<0.05$). The effect size refers to the minimum amount of difference that is clinically significant. According to the effect size values, there was no significant difference between time groups in the following parameters: Alb, ALT, AST, Pi, Ca, TC, Cr, Fe, glucose, BUN, lipase, TG, ALP, HDL-C, and CK ($ţ<0.30$).

Conclusion: When considering all of the results, nonfasting screening would not only be acceptable but also make physiologic sense.

Keywords: Clinical chemistry tests, database management system, statistical data analysis

An organism needs energy regularly for maintaining its life and functions. The main energy nutrients include carbohydrates, lipids, and proteins [1]. Fasting and nonfasting statuses are the most interesting issues in the nutritional area [2]. People mostly live in the nonfasting state during a normal 24-h cycle. Therefore, nonfasting metabolic panels may be a confident indicator of average biochemical parameter levels because the fasting state only consists after fasting for at least 8 h [3, 4].

At hospital laboratories, biochemical tests are generally performed on samples taken on fasting (preferably) between 08:00 and 10:00 [5]. Due to the increased workload of hospitals since the 2000s, patients have to wait for hours for sample collection even if they arrive at hospitals early in the morning [6]. This situation might cause stress in patients. Due to reasons such as the difficulties of employees in getting permission from their jobs and the fast pace of life, patients visit hospitals to provide their samples for testing in the evening. Additionally, the number of blood samples taken during nonfasting may often be higher than the number of those taken during fasting. In another aspect, it might not be possible for children and infants, who constitute an important part of the population, to stay hungry for many hours. The increased average age of the population also increases the number of patients aged over 70 years and those with chronic diseases. Prolonged fasting may cause health problems in these people. In fact, the body's metabolism is in a state of nonfasting

Address for correspondence: Abdullah Arpaci, MD. Department of Medical Biochemistry, Mustafa Kemal University Faculty of Medicine, Hatay, Turkey
Phone: +90 533 276 61 50 **E-mail:** arpaci57@gmail.com **ORCID:** 0000-0002-6077-8258
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for most of the day, except for times of compulsory fasting [6, 7]. Since 2010, some biochemical parameters such as the lipid profile have been routinely measured in nonfasting states in different countries [6, 8]. However, there is no study conducted in Turkey about the effects of fasting and nonfasting on biochemical parameters. In this respect, it can be thought that our study can present a different approach to the literature data.

Studies on the difference between the biochemical values of fasting and nonfasting states are generally related to lipids. So far, only a limited number of studies have reported measurements of other biochemical analytes including albumin (Alb), alkaline phosphatase (ALP), bilirubin, and uric acid [8, 9]. In this study, we evaluated a total of 18 biochemical parameters between 2010 and 2019, which includes Alb, aspartate aminotransaminase (AST), alanine aminotransaminase (ALT), Pİ, calcium (Ca), iron (Fe), gamma-glutamyltransferase (GGT), glucose, blood urea nitrogen (BUN), triglyceride (TG), ALP, creatine kinase (CK), total protein (TP), creatinine (Cr), high-density lipoprotein cholesterol (HDL-C), total cholestrol (TC), amylase, and lipase. The aim of this study is to examine the levels of these analytes during different hours of the day and compare them statistically according to time groups. The results of this study will contribute to other studies on this topic.

Materials and Methods

This is a retrospective study in which the data were obtained from the laboratory information system. Ethical approval for this study was obtained from the HMKU ethical committee (approval date is February 27, 2020, and the permission of ethics document's number is 07). The groups of the study were formed using the automation data of 202,235 patients who visited the outpatient clinics between January 1, 2010, and December 31, 2019. The data of patients from the inpatient clinics, intensive care units, oncology, pediatric, emergency service, and nephrology clinics were not evaluated to minimize the effect of chronic diseases on biochemical values and not considered to represent a healthy population. A total of 18 different biochemical parameters, which are most commonly requested by clinicians, were included in the study.

The patient blood samples were divided into eight groups starting from those drawn in the morning toward those taken in the afternoon as follows: (a) 07:00-07:59, (b) 08:00-08:59, (c) 09:00-09:59, (d) 10:00-10:59, (e) 11:00-11:59, (f) 12:00-13:59, (g) 14:00-14:59, and (h) 15:00-17:00 (a-h represent specified time groups). The blood samples delivered to the laboratory through a pneumatic system were centrifuged at 4000 rpm for 10 min after being kept for 20 min. Samples were analyzed using the original reagents and the same method using the Abbott Architect c8000 autoanalyzer from 2010 to 2017 and the Siemens Advia 1800 Clinical Chemistry System autoanalyzer from 2018 to 2019. During this process, the patient samples were analyzed after the internal quality control samples were tested daily at two levels and at 12-h intervals.

Continuous variables are shown as mean±standard deviation (SD) or minimum (min) value and maximum (max) value. Categorical variables are expressed as a number (n). Statistical analyses were performed using SPSS software (version 21, SPSS, Inc., Chicago, IL, USA). A p-value less than 0.05 was considered statistically significant in the evaluation of the differences between the groups. As clinical significance was investigated in this study, the effect size was used as another measurement parameter in evaluating the difference between the time groups [10]. The effect size refers to the minimum amount of difference that is clinically significant. The effect size value, which can produce more accurate results for a large number of patients, was used together with the p-value to evaluate the difference between groups.

According to Hedges' g statistic, a value of 0.2 is considered to be a small effect size, a value between 0.5 and 0.8 represents a medium effect size, and a value greater than 0.8 means a large effect size [11, 12]. In this study, the effect size values ranged between 0.10 and 0.30, t values recategorized as t=0-0.10, tt= 0.10-0.20, and ttt=0.20-0.30.

First, the data were cleaned out of noisy data, and outlier values were eliminated by using a box plot method. Then, one-way ANOVA was used to determine the statistical significance of the overall time periods. Brown-Forsythe analysis was used for homogeneity of variances in the time periods. Pairwise comparison of the time periods with statistically significant differences between variables was performed using Scheffe's and the Games-Howell post hoc tests. Finally, the effect size values were calculated separately for each variable and time period to determine the clinical significance level in the pairwise comparison of the time periods.

Results

A total of 202,235 patients were included in the study, the mean age of patients was 48.2±18.1 years, and the minimum and maximum values were 18 and 72, respectively. Of them, 114,961 (56.8%) were females with a mean age of 47.4±17.6 years and 87,274 (43.2%) were males with a mean age of 49.4±18.6 years.

Table 1 presents the comparison of the biochemical parameters according to the time periods of the day. We divided our samples into eight groups according to their collection times. As shown in Table 1, a statistically significant difference was observed for the Alb, ALT, AST, inorganic phosphorus (P_i), Ca, TC, Cr, Fe, glucose, BUN, lipase, TG, ALP, HDL-C, and CK parameters (Alb, p=006; ALT, p=0.004; ALP, p=0.011; AST, P_i, Ca, TC, Cr, Fe, glucose, BUN, lipase, TG, HDL-C, and CK, p<0.001). However, there was no statistically significant difference between the groups in terms of the amylase (p=0.661), GGT (p=0.233), and TP (p=0.066) parameters. All effect size values calculated to determine the clinical significance level in the pairwise comparisons of the groups were found to be lower than 0.30 (t<0.30). Accordingly, the changes in the Alb, ALT, AST, P_i, Ca, TC, Cr, Fe, glucose, BUN, lipase, TG, ALP, HDL-C, and CK parameters in time

Table 1. Comparison of biochemical parameters with time groups

Parameter	Units	07:00-07:59 (a) Mean±SD Min-Max	08:00-08:59 (b) Mean±SD Min-Max	09:00-09:59 (c) Mean±SD Min-Max	10:00-10:59 (d) Mean±SD Min-Max	11:00-11:59 (e) Mean±SD Min-Max	12:00-13:59(f) Mean±SD Min-Max	14:00-14:59 (g) Mean±SD Min-Max	15:00-17:00 (h) Mean±SD Min-Max	p	t
Alb	g/dL	4.26±0.42 2.5-5.7 n=4029	4.24±0.43 2.5-5.8 n=13772	4.25±0.44 2.44-5.8 n=11517	4.25±0.47 2.43-5.8 n=7370	4.25±0.48 2.43-5.7 n=3555	4.27±0.48 2.46-5.9 n=3710	4.25±0.49 2.5-5.8 n=2002	4.22±0.49 2.5-5.6 n=1367	0.006	<0.30
Amylase	U/L	68.3±27.4 13-176 n=765	67.1±26.6 9-175 n=3007	66.8±26 7-174 n=2706	66.6±27.1 8-173 n=1671	66±26.1 8-175 n=882	67.7±25.8 15-169 n=1252	67.9±27.2 19-169 n=537	66.8±25.8 9-175 n=368	0.661	-
AST	U/L	20.7±6.81 1-49 n=10012	21.3±6.99 2-49 n=35153	21.4±7.05 3-49 n=28066	21.5±7.22 1-49.1 n=17975	21.5±7.21 2.1-49.1 n=8537	21.4±7.18 3-49 n=10703	21.5±7.2 2.3-49 n=5940	21.5±7.06 2-49 n=4000	0.001	<0.30
ALT	U/L	20.2±10 2-60 n=13831	20.3±10.1 1-60.9 n=46863	20.4±10.3 1-60.8 n=37074	20.3±10.4 1-60 n=23597	20.1±10.3 2-60 n=11178	20.3±10.3 1-60 n=13917	20.7±10.7 1-60 n=7800	20.5±10.4 2.23-60 n=5159	0.004	<0.30
P_i	mg/dL	3.46±0.67 1.2-6.2 n=1175	3.43±0.65 1-6.3 n=4784	3.43±0.64 0.8-6.23 n=4098	3.46±0.66 1.1-6.3 n=2459	3.45±0.68 0.95-6.3 n=1051	3.56±0.67 1.2-6.1 n=1214	3.54±0.67 0.95-6.2 n=742	3.62±0.7 1.1-6.1 n=548	0.001	<0.30
Ca	mg/dL	9.32±0.53 7.05-11.5 n=4547	9.31±0.52 7-11.6 n=14706	9.32±0.53 7.03-11.6 n=12091	9.32±0.53 7.03-11.5 n=7415	9.32±0.57 7.1-11.6 n=3425	9.36±0.54 7.3-11.5 n=3673	9.32±0.55 7.2-11.6 n=2013	9.3±0.55 7.09-11.6 n=1445	0.001	<0.30
TC	mg/dL	193±43 67-383 n=3707	193±43.1 64-386 n=14093	193±43.1 64-386 n=10549	190±42.6 63-386 n=6071	191±44.2 67-380 n=2744	190±44 65.7-366 n=2581	192±46.5 71-383 n=1762	194±44.2 70-376 n=1371	0.001	<0.30
Cr	mg/dL	0.75±0.17 0.2-1.46 n=14920	0.75±0.18 0.2-1.46 n=51090	0.75±0.18 0.2-1.46 n=40109	0.76±0.18 0.2-1.46 n=25372	0.76±0.18 0.2-1.46 n=12142	0.77±0.18 0.2-1.46 n=14830	0.77±0.18 0.2-1.46 n=8255	0.77±0.18 0.2-1.46 n=5490	0.001	<0.30
Fe	µg/dL	65±38.8 2-245 n=2570	65.4±38.2 2-247 n=8338	67.6±39.3 2-247 n=6817	67.9±41 3-243 n=4311	65.8±39.5 3-243 n=2018	65.3±39.1 2-243 n=1867	63.6±37.9 5-206 n=1402	63.2±37.7 4-233 n=1091	0.001	<0.30
GGT	U/L	21.1±11.9 2-64 n=2250	20.3±11.7 1-64 n=7979	20.5±11.7 1-64 n=7084	20.4±11.8 1-64 n=4638	20.6±12.1 2-64 n=2374	20.4±11.5 2-64 n=4161	20.5±12 1-64 n=2244	20.2±11.5 2-64 n=1383	0.233	-
Glucose	mg/dL	95.5±16.2 42-151 n=11673	94.8±15.7 40-151 n=39901	93.9±15.5 41.5-151 n=31169	93.5±15.6 40-151 n=19303	93.2±15.9 42.2-151 n=9177	94.6±16.3 41-151 n=11391	94.4±15.9 41-151 n=6372	94.6±15.7 44.7-151 n=4457	0.001	<0.30
BUN	mg/dL	13±5.14 2-33.5 n=7929	13±5.26 2-33.6 n=28392	13±5.22 2-33.6 n=23402	13.1±5.28 2-33.6 n=15142	13.1±5.23 2-33.4 n=7694	13.5±5.26 2-33.6 n=10576	13.5±5.17 2-33.6 n=5806	13.4±5.03 2-33.5 n=3898	0.001	<0.30
Lipase	U/L	30.1±14.5 4-87 n=695	30.3±14.7 3-88 n=2739	29.6±14.7 3-88 n=2482	29±14.6 4-88 n=1491	29.1±14.7 4-85 n=786	31±14.9 2-88 n=1086	29.4±13.6 4-86 n=450	31.8±15 4-88 n=335	0.001	<0.30
TP	g/dL	7.24±0.58 5.04-9.5 n=2748	7.25±0.58 4.9-9.6 n=8373	7.26±0.59 4.84-9.6 n=6839	7.27±0.59 4.81-9.6 n=4340	7.23±0.61 4.85-9.4 n=2093	7.26±0.6 4.86-9.4 n=2407	7.27±0.6 5.2-9.6 n=1251	7.22±0.62 4.9-9.6 n=924	0.066	-
TG	mg/dL	146±78.7 23.1-464 n=4580	143±77.8 21-464 n=16262	141±77.6 23-464 n=12170	141±79.4 22-464 n=6968	148±83.2 22-464 n=3088	151±85 22-460 n=2896	147±85.7 25-464 n=1888	143±79.9 21-463 n=1518	0.001	<0.30

Table 1. Cont.

Parameter	Units	07:00-07:59 (a) Mean±SD Min-Max	08:00-08:59 (b) Mean±SD Min-Max	09:00-09:59 (c) Mean±SD Min-Max	10:00-10:59 (d) Mean±SD Min-Max	11:00-11:59 (e) Mean±SD Min-Max	12:00-13:59(f) Mean±SD Min-Max	14:00-14:59 (g) Mean±SD Min-Max	15:00-17:00 (h) Mean±SD Min-Max	p	t
ALP	U/L	75.4±26.6 24-179 n=2103	73.9±26.1 18-180 n=7796	74.1±26.2 18-180 n=6639	74.9±27.2 20-180 n=4221	75.3±26.1 19-179 n=1991	73.9±25.2 17-179 n=2376	72.4±25.3 23-179 n=1222	74.6±27.7 25-174 n=811	0.011	<0.30
HDL-C	mg/dL	44.1±11.5 14.3-94 n=3604	42.9±11.5 14.3-95 n=13770	43.2±11.6 14.5-95 n=10263	42.9±11.5 15.2-93 n=5919	42.4±11.9 14.8-95.1 n=2669	43.3±11.8 14-93 n=2481	42.9±12 15-92.1 n=1659	43.2±12.1 14.6-93.2 n=1344	0.001	<0.30
CK	U/L	88.7±46.4 16-274 n=1328	91.9±47.7 14-278 n=3179	91.4±47.9 14-279 n=2256	91.3±45.9 14-277 n=1447	93.8±49.9 16-276 n=618	100±52.2 16.9-266 n=725	101±53.1 24-270 n=394	97.3±53.5 21-275 n=322	0.001	<0.30

Alb: Albumin; AST: Aspartate aminotransaminase; ALT: Alanine aminotransaminase; P_i: Inorganic phosphorus; Ca: Calcium; TC: Total cholesterol; Cr: Creatinine; Fe: Iron; GGT: Gamma-glutamyltransferase; BUN: Blood urea nitrogen; TP: Total protein; TG: Triglyceride; ALP: Alkaline phosphatase; HDL-C: High-density lipoprotein cholesterol; CK: Creatine kinase.

groups were found not to be clinically significant (t<0.20). The effect size value was not found to be high even for the parameters with the highest differences (P_i and CK) (t<0.30).

Table 1 also shows the mean±SD and min-max values of the parameters. As shown in Table 1, the most noticeable change was observed in the TG level for the lipid panel (TC, TG, and HDL-C). The TG levels increased after 10:00-10:59, whereas this increase was statistically significant (p<0.001), but it was clinically weak significance (t=0-0.10). Regarding the kidney function tests of BUN and Cr, the lowest and highest levels were, respectively, 0.75±0.17 and 0.77±0.18 mg/dL for Cr, where the highest time-based change of mean concentration was 0.02 mg/dL. The levels throughout the time periods varied from 13.03±5.14 to 13.5±5.26 mg/dL for BUN, where the highest change of concentration was 0.54 mg/dL. These mean concentration changes expressed clinically weak significance (t=0-0.20). The minimum and maximum values were measured, respectively, as 2 and 247 µg/dL for serum Fe, which is frequently affected by diurnal variation during the day. However, the mean values of the patients were very close to each other, where the highest difference was 4.76 µg/dL. The highest mean value of serum Fe was measured between 10:00 and 10:59 (67.9±41 µg/dL), and the lowest mean value was measured between 15:00 and 17:00 (63.2±37.7 µg/dL). The difference between the groups in terms of serum glucose, which is an important metabolic indicator, was statistically significant (p<0.001), but it was clinically weak significant according to the effect size value obtained from the pairwise comparisons of the groups (t=0-0.20). The highest change of mean concentration in the serum glucose levels, for which different reference intervals were used according to the state of fasting and nonfasting at our laboratory, was found to be 2.26 mg/dL in time groups (t<0.30) (p<0.001).

Figure 1 presents the concentration-time graphs showing time-based changes in the biochemical parameters. These changes in the time periods are expressed in the graph as mean±SD. Thus, different groups are shown according to the post hoc tests, which express statistical and clinical significance between the time groups (a-h). As shown in Figure 1, the t value varied between 0 and 0.10 for Alb, BUN, ALT, lipase, and TC, between 0.10 and 0.20 for Cr, Ca, AST, Fe, ALP, glucose, TG, and HDL-C, and between 0.20 and 0.30 for P_i and CK, suggesting time-based changes in the parameters that are not clinically significant. As there was no statistically significant difference between the groups in terms of GGT, amylase, and TP at any time period, the t value was not calculated.

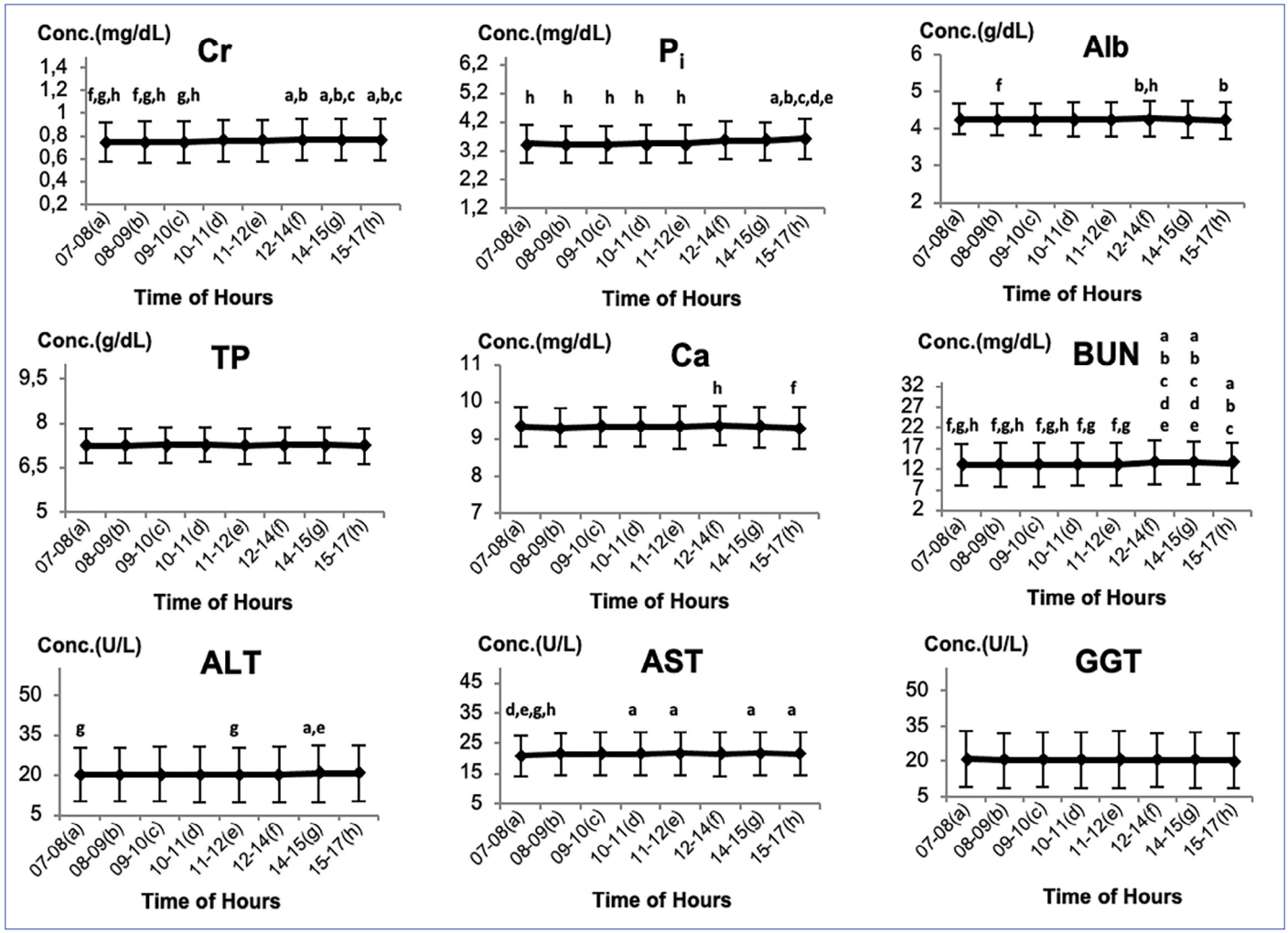
Discussion

Laboratory tests, which are the main sources of medical data, are an important part of modern medicine and quality healthcare services, and laboratories are very important health centers for clinicians to evaluate patient diagnosis, treatment, and follow-up. The effect of clinical laboratories in deciding diagnosis and treatment is approximately 70% [13]. One of the most

important factors for reliable laboratory results is to ensure the appropriate test request and optimal sample collection [14]. The body metabolism is in a state of nonfasting for most of the day, except for times of compulsory fasting, and many European countries have changed their reference intervals for biochemical parameters according to nonfasting values [6].

In the literature approach to the lipid profile, which is adopted in many countries such as the USA, Switzerland, and Canada, there is no significant difference between fasting and nonfasting status, and thereby many countries make changes to their reference intervals [5, 15]. A recent prospective study by Wang et al. [16] evaluated lipid profile values in fasting and 4-h postprandial status. The results showed that there were no significant differences in lipid levels between the fasting and nonfasting samples. In our study, although there was a statistically significant difference between the time groups for HDL-C (n=41,709), TC (n=42,878), and TG (n=49,370) tests ($p<0.001$), this difference was not clinically significant according to the effect size values that were calculated ($t<0.30$). Additionally, the lowest and highest mean values for TG were 141±7.62 and 151±85 mg/dL, respectively, where the highest change of mean value was 10.3 mg/dL. The mean values for HDL-C and TC were very close to each other, where the highest differences between the mean values were 1.69 and 3.4 mg/dL, respectively (Table 1). Several studies have shown that fasting is not substantially necessary for routine lipid level measurements [3, 16, 17]. As a result of studies including children, women, and men with diabetes from the USA, Denmark, and Canada, plasma lipids and lipoproteins have been observed to change slightly in response to the patient's usual food intake ($p>0.05$). These studies, which compared fasting and nonfasting lipid profiles, found no significant difference in HDL-C concentrations, despite minor decreases in plasma TG, TC, and LDL-C concentrations ($p>0.05$). As a result, it has been reported that these minimal and temporary changes in lipid concentrations are not clinically significant [6, 8, 18]. Similarly, the comparison of the mean±SD values of lipid parameters was not clinically significant ($t<0.30$) in our study and is consistent with the data reported in the literature (Table 1).

American Heart Association and the British National Institute of Health and Care have recommended the use of nonfasting lipid profiles. For that reason, many laboratories in the USA and the UK have revised their reference intervals [19, 20]. Indeed, the Hypertension Canada's Guidelines, the European Society



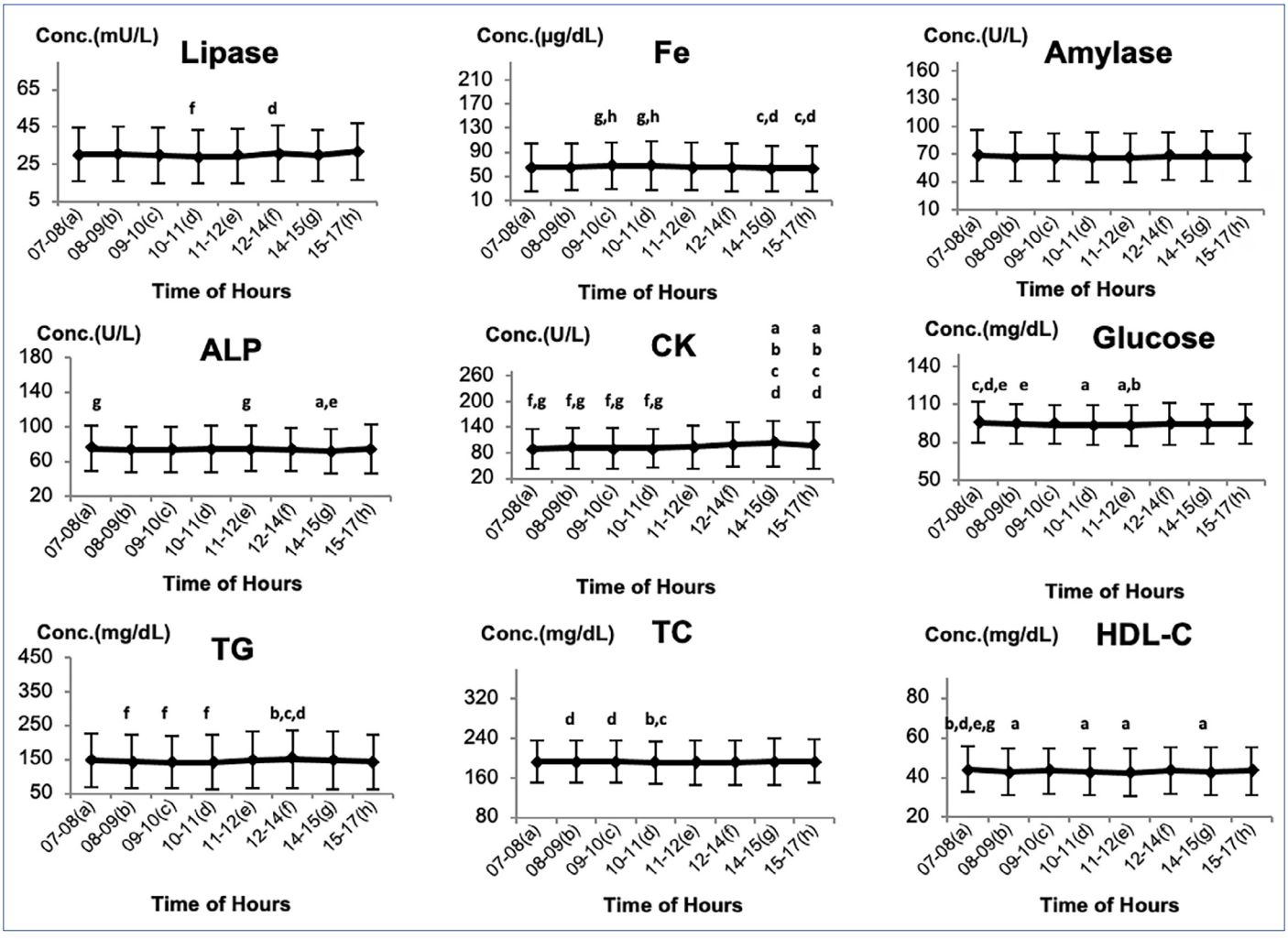


Figure 1. Evaluation of clinical significance of biochemical tests with the effect size.

Alb, BUN, ALT, lipase, TC: t; Cr, Ca, AST, Fe, ALP, glucose, TG, HDL-C: tt; P_i, CK: ttt; GGT, amylase, TP: no difference. t=0-0.10; tt=0.10–0.20; ttt=0.20–0.30. Statistically different time groups are shown in lowercase alphabets (a-h). Cr: Creatinine; P_i: Inorganic phosphorus; Alb: Albumin; TP: Total protein; Ca: Calcium; BUN: Blood urea nitrogen; ALT: Alanine aminotransaminase; AST: Aspartate aminotransaminase; GGT: Gamma-glutamyltransferase; Fe: Iron; ALP: Alkaline phosphatase; CK: Creatine kinase; TG: Triglyceride; TC: Total cholesterol; HDL-C: High-density lipoprotein cholesterol.

of Cardiology, the European Atherosclerosis Society, and relevant organizations in many other countries reviewed their laboratory guidelines according to nonfasting status [19, 21].

Pasic et al. [9] have collected four blood samples from each of 27 healthy individuals 2 h after breakfast, 2 h after lunch, and in the afternoon (at 17:00), and they compared laboratory analytes including ALT, AST, ALP, GGT, TP, Ca, glucose, TG, HDL-C, CK, Cr, amylase, Fe, total bilirubin (TBIL), lactate dehydrogenase, P_i, and transferrin, which are thought to be affected in fasting status. As a result, they found a significant difference between the fasting and nonfasting levels of Alb, ALT, AST, ALP, Ca, TP, and TG ($p<0.05$), and they observed diurnal variation in Alb, ALP, Fe, ALT, AST, Ca, TC, and HDL-C. In our study, a statistically significant difference was found between the groups in terms of all parameters except amylase, GGT, and TP ($p<0.05$), but this difference was not clinically significant (t<0.30). As shown in Table 1 and Figure 1, the mean concentration changes in the parameters were very close to one another. Moreover, this study found the lowest and highest mean concentrations as, respectively, 20.1±10.3 and 20.7±10.7 U/L for ALT and 20.7±6.81 and 21.5±7.2 U/L for AST. This change was below 1 U/L, suggesting that collecting blood samples at any time of the day does not constitute a significant difference for ALT and AST (t=0-0.20). Some studies on fasting and nonfasting evaluation of the ALT and AST parameters suggested no significant difference between these two metabolic states, whereas others suggested a significant difference between them [22, 23].

There is a limited number of studies suggesting no significant difference between the status of fasting and nonfasting for ALP [24, 25]. In this study, we evaluated a total of 27,159 ALP samples and found that the mean±SD values between the time periods were very close to one another, where the lowest and highest mean values were 72.4±25.3 and 75.4±26.6 U/L, respectively (Table 1). Plumelle et al. [24] conducted a study with a total of 20 healthy adults aged between 23 and 33 years.

They evaluated 37 biochemical parameters in fasting and nonfasting, using blood samples collected in three groups: 12-h fasting, 3 h after breakfast, and 3 h after lunch. They found that only 7 out of 37 biochemical parameters were affected in fasting and nonfasting status (uric acid, TBIL, brain natriuretic peptide, Cr, glucose, P, and TP), where the difference was not clinically significant because the total change limit (TCL) was observed the same [$TCL=\sqrt{(2.77CVa)^2+(0.5CVb)^2}$].

The findings from this study suggest that there is no clinically significant difference in all biochemical parameters except GGT, amylase, and TP according to the effect size values. Studies on the difference between the biochemical values of people in fasting and nonfasting states are generally conducted about lipids. The present study confirms previous findings and contributes to research that measures biochemical parameters at different time periods.

Limitations of the study

This is a retrospective study in which 202,235 patients were evaluated and the first application of each patient was accepted. Planning the study prospectively may lead to a reduction in the data set and a subjective assessment that requires survey research based on the collection of information from patients. In addition, direct measurement of LDL-C is an expensive, time-consuming technique and requires large sample volumes. Also, we did not evaluate the association between levels of direct LDL-C and estimated LDL-C. Therefore, we excluded both types of LDL-C from the study. Although our study was designed as a study evaluating biochemical parameters at different hours of the day, it would be better to evaluate biological variations or diurnal variations. In future studies, it may be planned to evaluate these variations together with biochemical parameters.

Conclusion

Consequently, we suggest that recording nonfasting samples routinely taken every hour at clinical laboratories, additionally, could be revised with minor changes in the reference intervals. When considering all of the findings from this study, nonfasting screening would not only be acceptable but also make physiologic sense. In recent years, there has been an increasing amount of literature on fasting and nonfasting issues, which has also increased the interest of researchers in the area. However, there are limited studies on this issue in Turkey. This research will serve as a base for future studies, and the findings from the study make contributions to the current literature.

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