The Evaluation of Blink Reflex in Patient with Prediabetes

Prediabetli Hastalarda Blink Refleksin Değerlendirilmesi

Erkan Cüre², Serker Kerbaş², Medine Cumhur Cüre³, Aynur Kerbaş³, Süleyman Yüce¹, Ahmet Tüfekçi², Bayram Kızkaya¹

¹ Recep Tayyip Erdoğan University, Department of Internal Medicine, Rize, Turkey
² Recep Tayyip Erdoğan University, Department of Neurology, Rize, Turkey
³Recep Tayyip Erdoğan University, Department of Biochemistry, Rize, Turkey

Abstract

Purpose: The aim of this study was to investigate the blink reflex (BR) which is related to V. and VII. cranial nerves and whether or not it slows down or not in patients with prediabetes. Materials and Methods: A total of 90 subjects were enrolled in the study, 60 patients (43 female, 17 male) with impaired fasting glucose and impaired glucose tolerance and 30 healthy controls (16 female, 14 male). Electromyography was performed in both groups. After electrical stimulation of the supraorbital nerve, two responses in the orbicularis ocular muscle occurred, the early ipsilateral response “R1” and the late bilateral response “R2” latency times were recorded. Results: The mean age of prediabetes and the control groups was 45±9 and 41±11 years respectively. The patients group were longer Early right ipsilateral response (p<0.001), Late right ipsilateral response (p<0.001), Late right contralateral response (p<0.001), Early left ipsilateral response (p<0.004), Late left ipsilateral response (p<0.001) and Late left contralateral response (p<0.015) than control group. Fasting plasma glucose and lipid parameters were significantly higher in the prediabetes group. Conclusion: There is a slowdown of BR in the prediabetes stage. The accompanying of hyperlipidemia contributes to the reduction of this reflex. The decrease of BR demonstrates the involvement of V. and VII. cranial nerves. By performing the BR, neuropathy can be detected at an early stage in patients with prediabetes.

Keywords: Blink reflex; neuropathy; prediabetes; impaired fasting glucose; impaired glucose tolerance
Introduction

Eye-blink is a voluntary or spontaneous reflex process that protects the eye. Blink reflex (BR) occurs as a result of supraorbital nerve stimulation by one sided superficial bipolar electrodes and by taking a two-sided record from orbicularis oculi muscles stimulated by superficial needle electrodes (1,2). BR is a beneficial electrodiagnostic method for the evaluation of neuropathies. This test evaluates the afferent arc of cranial nerve V, efferent arc of cranial nerve VII, pons, and lateral medulla (3). It may be compared clinically with the corneal reflex. This reflex may be affected by facial paralysis, trigeminal nerve lesions, brainstem and spinal cord lesions (3-7).

As it is known in this era, diabetes is commonly seen and its prevalence is increasing worldwide. The incidence of prediabetes (intermediate hyperglycemia) is increasing steadily with increased obesity, a change in nutritional intake with a predominance of fast food and reduced exercise. Diet, exercise and metformin/acarbose are used in patients with prediabetes to slow down progression to diabetes (8,9). Without medical treatment or changing life style, the progression to diabetes is accelerated (10). In this condition, exposure to hyperglycemia for a long time increases the incidence of neuropathy. Large studies have shown that 5–14% of diabetic patients >40 years old have neuropathy (11,12). Previously studies have displayed at about 40% prevalence of impaired glucose tolerance (IGT) among subjects with idiopathic neuropathy (13,15), compared with some 26% in the age-matched broader population (16).

The neuropathy associated with IGT is predominantly sensory, painful and characterized by small-fiber injury (17). Recently impaired fasting glucose (IFG) and IGT were reported to cause neuropathy via small fiber injury (13,18). Skin biopsy with intraepidermal nerve fiber density measurement is a very specific method for the determination of neuropathy (15,19). In contrast to this test to demonstrate the involvement of cranial nerves, the BR is quite a simple and uncomplicated test. This reflex is known to be decreased in diabetic patients. If we can demonstrate the slowing down of the BR in patients with prediabetes, we can generalize it is usage in clinics to demonstrate, and follow up on, the progression of neuropathy. The aim of this study was to determine the slowing down of the BR in patients with prediabetes.

Materials and Methods

Patients

Patients with history of neuropathy, diabetes, $B_{12}$ deficiency, thyroid disease, pregnancy, chronic renal failure, acute or chronic infection, use of drugs that may cause hyperglycemia, hypertension or pre-hypertension (systolic blood pressure 135-139 mmHg, diastolic blood pressure 85-89 mmHg) and any other known chronic disease were excluded from the study. Subjects who did not have any of the exclusion criteria diseases, fasting plasma glucose (FPG) within 100-126 mg/dl, not receiving any medical treatment diagnosed with IFG or IGT after performing oral glucose tolerance test (OGTT) and were included in the study group. The American Diabetes Association has defined a “prediabetes” state of either IFG or IGT 2 hours after the administration of 75 g of oral dextrose. A FPG of 100 mg/dL to 125 mg/dL is defined as IFG and plasma glucose 2 hours after a 75 g anhydrous dextrose oral load of 140 mg/dL to 199 mg/dL is defined as IGT (20).

A total of 60 patients, 43 female and 17 male, were included in the patients group. Patients who had received a diagnosis of prediabetes within the last 1-12 months were selected (median 3 month). Healthy subjects who did not have any known disease and having FPG less than 100 mg/dL were selected for the control group. There were 16 female and 14 male included in the control group. The study was approved by the local ethics committees (Approval numbers: 2012/128).

To obtain BR by electromyography

The design of the study is created from a similar model of a previous study (21). The electromyography (EMG) recording in standby, patients with prediabetes and the control group subjects were supine on a bed in a warm room with their eyes slightly closed. The left and right supraorbital nerves were stimulated percutaneous with bipolar surface electrodes after putting them above the eyebrow in the region where the supraorbital nerve emerges from the skull. The reflex responses were recorded by using two surface electrodes placed on the cheekbone above the orbicularis oculi muscle in line with the pupil in forward gaze to record the response of the muscle. The EMG signal was then conducted to the recording device. The reference electrode was placed on the lateral surface of the nose and the ground electrode was placed on the arm that is electrically inactive site.

The amplitude of a blink’s EMG rarely exceeds few hundred microvolts; so recording conditions should improve the flow of current from the skin surface to the electrodes. To reduce the impedance between skin and electrode gel, skin is prepared by removing makeup and
dead skin cells. After that, the electrode gel is massaged to form a thin layer on the recording place. The electrical stimulation of the supraorbital nerve reveals two responses in the orbicularis oculi muscle, the early ipsilateral response “R1” and the late bilateral “R2” responses. Sample of BR in patients with prediabetes are shown figure 1.

The duration of the stimulus was 0.1-0.2 ms and its intensity was set to a 100-microvolt/division and always under the pain threshold, in order to evoke early R1 and late R2 responses and simultaneously to avoid any activation of nociceptive afferents. The signals of EMG were amplified with a frequency response between 20 Hz to 3 kHz, which allowed accurate analyses of short latency responses. The latency times of R1 and R2 reflex responses were measured from the stimulus artifact to the initial response of the orbicularis oculi muscle. No auditory or visual prepulse stimulation was given to the subjects.

Statistics and ethical issues

The results were reported as the mean ± SD. The data analysis was performed using the statistical software SPSS for Windows (version 13.1; SPSS, Chicago, IL, USA). All the results were analyzed by applying the Kolmogorov-Smirnov for the determination of normal and abnormal data distribution. The statistical significance of the differences in all parameters between the prediabetes and the control group were analyzed using the independent sample t-test and Man Whitney U test. The differences were considered significant at P < 0.05.

Results

The mean age of prediabetes and the control groups was 45±9 and 41±11 years respectively. According to OGTT results, there were IFG group with 45 patients and IGT group with 15 patients. When both groups were compared regarding EMG findings, the results were similar. In comparison with the control group, the patients group had early right ipsilateral response (RR1) (p<0.001), late right ipsilateral response (RR2i) (p<0.001), late right contralateral response (RR2c) (p<0.001), early left ipsilateral response (LR1) (p<0.004), late left ipsilateral response (LR2i) (p<0.001) and late left contralateral response (LR2c) (p<0.015).

Glucose and lipid parameters were significantly higher in the prediabetes group. The demographic characteristics and the results of biochemical parameters of the patients are shown in table 1. EMG findings are shown in table 2.

Discussion

In this study, IFG and IGT patients (patients who are not diabetic but exposed to hyperglycemia) were included. According to the results of this study, the BR of non-diabetic patients with FPG >100 mg/dL has been found to be lower than healthy patients with FPG < 100 mg/dL. Body mass index (BMI), waist/hip ratio and the age of the two groups were selected to be similar to achieve the least effect on the results. The levels of total cholesterol (TC), triglyceride (TG) and low density lipoprotein (LDL) were found to be significantly higher in the prediabetes group. However, these levels were found to be within normal range in the two groups. The level of alanine aminotransferase (ALT) was found to be higher in the prediabetes group than the normal group.

According to the results of this study in patients with prediabetes showed that early and late phase of BR were obviously lower compared to the control group. In diabetic patients, it is known that the III. V. and VII. cranial nerves are frequently affected (22). A study conducted on patients with prediabetes has shown that these nerves were affected in a similar fashion to diabetic patients (17). In this study, non-diabetic patients with moderate hyperglycemia were found to have a lower bilateral BR. A reduced BR in patients with prediabetes demonstrates the development of neuropathy so this may be used as a screening test.

Neuropathy is one of the late complications of diabetes; it appears years after the diagnosis has been made. There have been few studies which used the BR to demonstrate the involvement of cranial nerves in diabetic patients (23-26). Nevertheless, there is evidence that neuropathy develops earlier in the course of hyperglycemia than other microvascular diabetic complications. In a wide investigation, more than 20% of early diabetic patients had physical examination or electrophysiological findings of neuropathy, while most of them had no proteinuria or retinopathy (27). In recent years, the development of neuropathy (especially small fiber neuropathy) in patients with prediabetes has been reported (28-31). It is more focused on the neuropathy of IGT. Divisova et al. have identified small fiber neuropathy in patients with prediabetes interval of one month to one year (31). This finding supports our study. In current study, subjects with less than one year’s duration of prediabetes were also included and EMG findings were similar in IFG and IGT groups.

It is still being debated whether prediabetes needs treatment or not. However, studies have reported that the stage of prediabetes, if not treated with lifestyle changes or medications, may progress quickly to diabetes (26). Moreover, the level of plasma glucose of
patients in prediabetes stage should be kept under strict control in order to prevent neuropathic complication. Our study also supports the thesis that BR slowdown demonstrates cranial nerves’ involvement during early prediabetes.

Patients with diabetes or prediabetes have cellular damage that ends in neuropathy due to hyperglycemia. As a result of raised glycolysis, generation of reactive oxygen radicals can lead to damage of nerves (32). In another mechanism as a result of increasing polyol pathway cellular osmolarity is increased consequently reducing the level of NADPH so oxidative stress is elevated. Eventually, increased flux through the hexosamine pathway is related to inflammatory injury (33,34). Increasing advanced glycation end products (AGE) is another pathway that participates in the development of neuropathy. The elevation of AGE damages structural cellular protein, lipid and nucleic acids. Inflammatory cascade is started by AGE so NADPH oxidase activation occurs and oxygen radicals are formed by oxidative stress and, as a result, neurons are damaged (35,36).

In current study, even though the levels of lipid were within the normal range suggested by the guidelines, the TC, TG and LDL levels of the prediabetes group were higher than the control group. There is relationship among dyslipidemia, diabetes and neuropathy (37). Previous studies have shown how LDL gives rise to oxidative stress and how it ends in apoptosis in neurons (38). Another mechanism is the direct toxic effect of free fatty acids on Schwann cells. Promoting the release of inflammatory cytokines from adipocytes and macrophages contributes to the damage (39). The contribution of both hyperglycemia and hyperlipidemia to neuropathy formation and the slowdown of the BR compared to the control group these two conditions may be formed by a synergistic effect. Most of the subjects’ BMI were between 25-30 kg/m², so they were slightly obese. Neuropathy was reported in subjects with metabolic syndrome (40). Thus obesity may have an effect on the slowdown of the BR. However, the prediabetes group and the control group had similar BMI so obesity has not affected the results of the study.

**Limitation of study**

In the study, the levels of insulin and HbA1c were not evaluated. The HbA1c levels of prediabetes patients are generally normal or close to normal so not being evaluated may not have an obvious effect on the study. Since OGTT was performed, HbA1c was not used in screening the patients. The evaluation of insulin levels could have an effect on the results of the study. Insulin resistance in obese patients, in particular, might have a correlation with BR. Our study has a pilot study feature so future studies may demonstrate the relationship among insulin, HbA1c levels and BR.

**Conclusion**

According to the results of the study BR was found to be delayed in both IFG and IGT patients groups. In prediabetes patients, by performing BR, the involvement of V. and VII. cranial nerves can be demonstrated. Thus, neuropathy can be detected at an early stage by performing BR which is a simple and uncomplicated test.

**Acknowledgments**

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**References**

Table 1. The main characteristics and laboratory parameters of the two groups.

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Patients</th>
<th>Controls</th>
<th>P values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex (M/F)</td>
<td>43/17</td>
<td>16/14</td>
<td>0.104</td>
</tr>
<tr>
<td>Age (years)</td>
<td>45±9</td>
<td>41±11</td>
<td>0.054</td>
</tr>
<tr>
<td>Waist Circumference (cm)</td>
<td>M 97.4±8.6</td>
<td>95.7±7.4</td>
<td>0.653</td>
</tr>
<tr>
<td></td>
<td>F 92.5±8.4</td>
<td>88.9±3.9</td>
<td>0.063</td>
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<tr>
<td>Waist/Hip ratio</td>
<td>0.93</td>
<td>0.91</td>
<td>0.659</td>
</tr>
<tr>
<td>BMI (kg/m^2)</td>
<td>29.8±4.0</td>
<td>28.6±3.0</td>
<td>0.115</td>
</tr>
<tr>
<td>FPG (mg/dL)</td>
<td>107±5</td>
<td>91±6</td>
<td>0.001</td>
</tr>
<tr>
<td>AST (IU/L)</td>
<td>19±6</td>
<td>18±4</td>
<td>0.421</td>
</tr>
<tr>
<td>ALT (IU/L)</td>
<td>21±8</td>
<td>16±5</td>
<td>0.001</td>
</tr>
<tr>
<td>BUN (mg/dL)</td>
<td>30±9</td>
<td>28±8</td>
<td>0.415</td>
</tr>
<tr>
<td>Creatinin (mg/dL)</td>
<td>0.7±0.1</td>
<td>0.8±0.1</td>
<td>0.073</td>
</tr>
<tr>
<td>TC (mg/dL)</td>
<td>201±37</td>
<td>169±33</td>
<td>0.001</td>
</tr>
<tr>
<td>TG (mg/dL)</td>
<td>132±63</td>
<td>87±34</td>
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</tr>
<tr>
<td>LDL (mg/dL)</td>
<td>128±31</td>
<td>107±21</td>
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</tr>
<tr>
<td>HDL (mg/dL)</td>
<td>46±10</td>
<td>44±9</td>
<td>0.470</td>
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ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, Body mass index; BUN, blood urea nitrogen; F, Female; FPG, fasting plasma glucose; HDL, High density lipoprotein. LDL, Low density lipoprotein; M, Male; TC, Total cholesterol; TG, Triglycerides.

Table 2. EMG findings of the prediabetes and control groups.

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Patients</th>
<th>Control</th>
<th>P values</th>
</tr>
</thead>
<tbody>
<tr>
<td>RR1 (ms)</td>
<td>11.5±1.22</td>
<td>10.4±1.37</td>
<td>0.001</td>
</tr>
<tr>
<td>RR2i (ms)</td>
<td>34.7±3.38</td>
<td>32.0±2.03</td>
<td>0.001</td>
</tr>
<tr>
<td>RR2c (ms)</td>
<td>34.9±3.07</td>
<td>32.7±2.53</td>
<td>0.001</td>
</tr>
<tr>
<td>LR1 (ms)</td>
<td>11.6±1.02</td>
<td>10.9±0.89</td>
<td>0.004</td>
</tr>
<tr>
<td>LR2i (ms)</td>
<td>34.7±3.10</td>
<td>32.1±1.86</td>
<td>0.001</td>
</tr>
<tr>
<td>LR2c (ms)</td>
<td>35.1±3.01</td>
<td>33.4±2.24</td>
<td>0.015</td>
</tr>
</tbody>
</table>

RR1, Early right ipsilateral response; RR2i, Late right ipsilateral response; RR2c, Late right contralateral response; LR1, Early left ipsilateral response; LR2i, Late left ipsilateral response; LR2c, Late left contralateral response.
Figure 1. BR pattern in one prediabetes patient's performed by EMG.

BR: Blink Reflex