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Original Article

Correlation of HbA1c and S. creatinine along with microbiological profiling of infected ulcers; cases of diabetic patients

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ABSTRACT

Statement of the problem: The rate of diabetes mellitus is increasing globally as a result of increased levels of blood glucose. The elevated blood glucose level results due to impairment in the production and action of insulin. An increase in glycated hemoglobin is associated with higher blood glucose levels that further results in nephropathy, neuropathy, retinopathy, and cardiovascular disease. Therefore, the study has investigated the correlation between HbA1c and creatinine levels among diabetic patients, suffering from foot ulcerations.

Methods: A prospective cohort hospital based-study has retrieved hospital records of patients during January 2009 and February 2015. A total of 192 patients, admitted in the Rajiv Gandhi Centre for Diabetes and Endocrinology, of Jawaharlal Nehru Medical College Hospital, Aligarh Muslim University, Aligarh, India were recruited.

Results: It has been examined that there is a positive correlation between ulcer duration and BMI, amputation rate and BMI, gender and BMI, hospital stay and BMI, HbA1c and BMI, Hb and BMI and triglyceride and BMI at 5% level of significance.

Conclusion: Findings have shown positive association with gender, diabetes duration, ulcer size, grade of ulcer, amputation rate, hospital stay, Hb, SGOT/AST and triglyceride.

1. Introduction

Foot ulcerations are amongst the major long-term complications of diabetes. These ulcerations are likely to result in gangrene, and cause amputation of lower extremities and mortality [1]. A study conducted by Li et al. [2] showed that approximately 25–90% of the amputations occurred among diabetic patients as compared to the normal patients. The most common complication of diabetes is foot ulceration, which is likely to affect 15% of the total population worldwide [3]. The severe condition of diabetic foot ulcerations may result in the hospitalization of patients. A study revealed that foot ulcerations among diabetic patients accounted for majority of the cased of non-traumatic amputation of the lower limbs [4]. As estimated, the number of diabetic cases is likely to raise to 366 million by the year 2025 [5].

The incidence of diabetes mellitus is known to increase globally as a result of increased levels of blood glucose. The blood glucose levels are likely to elevate as a result of impairment in the production and action of insulin. An increase in glycated hemoglobin is directly linked to high blood glucose levels, and also results in nephropathy, neuropathy, retinopathy, and cardiovascular disease. A kidney disease known as diabetic nephropathy is caused by high levels of blood glucose and high blood pressure and is the leading cause of kidney failure in United States. Diabetic nephropathy and diabetic retinopathy can lead to systemic and visual morbidity and mortality. This cannot only result in blindness and end stage renal disease, but can impose huge economic, social and medical costs to

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the patient as well as the health care system [6].

The physicians consider elevated HbA1c as a significant indicator of diabetes mellitus. The normal range of HbA1c among non-diabetic patients is 4–5.9%. Among patients with controlled diabetes, HbA1c is less than 7% and patients with poorly controlled diabetes has HbA1c level above 8% [2]. Apart from HbA1c, creatinine is considered as a reliable indicator of kidney functioning among diabetic patients. Any dysfunction of the kidneys leads to an increase in the creatinine level in blood. Serum creatinine levels have to be constantly monitored to prevent the progression of diabetic kidney disease. Therefore, the association of HbA1c and serum creatinine levels among diabetic patients with foot ulcerations have been considered significant in this study. The study has also investigated the correlation between HbA1c and creatinine levels among diabetic patients, suffering from foot ulcerations.

2. Methodology

A prospective cohort hospital based-study has been conducted, and hospital records of patients during the time period of January 2009 and February 2015 were retrieved. A total of 192 patients, admitted in the Rajiv Gandhi Centre for Diabetes and Endocrinology, of Jawaharlal Nehru Medical College Hospital, Aligarh Muslim University, Aligarh, India have been recruited on the basis of their availability at the hospital. The study was carried out in accordance with principles of the Declaration of Helsinki as revised in 2001, and subsequent consent of the patients was taken to participate in the study. The patients, who had been suffering from serious chronic diseases, inflammatory or infectious diseases, autoimmune and rheumatic diseases, cancer, haematological diseases, severe renal or liver failure, or under treatment with anti-inflammatory drugs, have been excluded. The demographic details of the patient included their ages and genders. In the first step, fasting blood sugar levels of the respondents have been evaluated. In the next step, the patient was given a glucose solution, and blood glucose levels have been checked after 2 h. Moreover, serum has been obtained through the technique for centrifugation, and it has been stored at −70°C. Blood has been taken to calculate glycosylated hemoglobin and serum creatinine levels. Method of culture of specimens, antimicrobial susceptibility testing was described elsewhere in previous studies [7]. Furthermore, the data obtained has been analyzed using descriptive statistics.

2.1. Ethical clearance

This study was approved Bio-Ethical Committee (BEC) of Faculty of Medicine, Aligarh Muslim University, Aligarh registered under Drug Controller General of India (DCGI), Government of India under registration Number: ECR/419/Inst/UP issued under Rule 122DD.

3. Results

A total of 192 patients participated in the study, out of which 112 were males and 80 were females. Patients who were equal to or less than 20 years were 1.4%, 21–40 years were 15.6% and 41–60 years were 54.8%. The findings have indicated that mostly patients had HbA1c greater than 6.9 level ($n = 183$). Moreover, results have shown a significant relationship between Grade of ulcer and HbA1c ($p = 0.068$) while the relationship between HbA1c and Hb was significant ($p = 0.004$) (Table 1).

Table 2 has shown the factors associated with BMI after adjustment. The findings have indicated that mostly patients (40.1%) belong to lean category followed by normal category (28.6%), obese category (16.7%) and 14.6% participants were overweight. Moreover, findings have shown that patients in lean category (29.9%) usually stay longer in the hospital followed by normal category (27.3%), obese category (18.8%) and over weight category (10.7%). Similarly, HbA1c was found in greater extent among lean category (96.1%) followed by normal category and obese category.

The factors associated with creatinine clearance after adjustment have been presented in Table 3. The findings have shown that there was a positive association between Category A patients and discharge status, hospital stay, HbA1c, WBC count, serum creatinine and SGPT/AST. Moreover, findings have indicated that Category B patients were associated with discharge status, hospital stay, HbA1c, WBC count, and SGPT/AST. In the context of Category B, findings have shown a positive association with ulcer size, discharge status, hospital stay, HbA1c, WBC count, Hb, serum creatinine, SGOT/AST, SGPT/AST and total cholesterol. In the context of Category C, findings have shown a positive association with ulcer size, discharge status, hospital stay, HbA1c, WBC count, Hb, serum creatinine, SGOT/AST, SGPT/AST and total cholesterol.

Table 4 has presented correlation between BMI, HbA1c and serum creatinine with clinical and laboratory variables. From the findings, it has been examined that there is a positive correlation between ulcer duration and BMI ($p = 0.041$), amputation rate and

Table 1
Baseline characteristic of participants.

	HbA1c < 6.9 N = 7		HbA1c ≥ 6.9 N = 183		P value
	N	%	N	%	
Male/Female	3/4	42.9/57.1	122/61	66.7/33.3	0.193
Age >40 years	7	100	134	73.2	0.112
Grade of ulcer (Wagner)					
1	3	42.9	50	27.3	0.068
2	0	0	68	37.2	
3	2	28.6	43	23.5	
4	0	0	11	6	
5	2	28.6	11	6	
WBC count ($10^3/\mu l$)	5	71.4	136	74.3	0.083
Hb (g/dl)	7	100	93	50.8	0.004
SGOT/AST (>34IU/L)	7	100	64	34.9	0.339
SGPT/AST (>35 IU/L)	7	100	94	51.3	0.462
LDL-C (>100 mg/dl)	7	100	109	59.5	0.430
Total cholesterol (>150 mg/dl)	2	28.6	95	51.9	0.430
HDL-C (<40 mg/dl)	0	0	48	26.2	0.465
Triglycerides (>200 mg/dl)	3	42.7	54	29.5	0.434

Table 2
Factors associated with BMI after adjustment.

	Obese [≥25 kg/m^2] N = 32	Over weight [23–24.9 kg/m^2] N = 28	Normal [18.5–22.9 kg/m^2] N = 55	Lean [<18.5 kg/m^2]N = 77
N				
Male/Female	32 (16.7%)	28 (14.6%)	55 (28.6%)	77 (40.1%)
Grade of ulcer (Wagner)				
1	10 (18.9%)	3 (5.7%)	20 (37.7%)	20 (37.7%)
2	3 (5.7%)	11 (16.2%)	6 (8.8%)	34 (50%)
3	5 (10.6%)	9 (19.1%)	19 (40.4%)	14 (29.8%)
4	0	5 (10.6%)	3 (5.7%)	3 (5.7%)
5	0	0	7 (53.8%)	6 (8.8%)
Hospital stay (>1month)	6 (18.8%)	3 (10.7%)	15 (27.3%)	23 (29.9%)
HbA1c (>7%)	30 (93.8%)	28 (100%)	51 (92.7%)	74 (96.1%)
WBC count (10^3/μl)	9.71 ± 4.17	8.68 ± 4.20	8.77 ± 4.12	11.19 ± 4.18
Hb (g/dl)	10.4 ± 2.15	9.92 ± 2.12	10.09 ± 2.14	10.64 ± 2.17
Serum creatinine (>1.5 mg/dl)	5 (15.6%)	8 (28.6%)	7 (12.7%)	18 (23.4%)
SGOT/AST (>34IU/L)	22 (68.7%)	19 (67.8%)	17 (30.9%)	13 (16.8%)
SGPT/AST (>35 IU/L)	17 (53.1%)	22 (78.5%)	41 (74.5%)	21 (27.2%)
LDL-C (>100 mg/dl)	31 (96.8%)	28 (100%)	26 (47.2%)	31 (40.2%)
Total cholesterol (>150 mg/dl)	19 (59.3%)	21 (75.0%)	41 (74.5%)	16 (20.7%)
HDL-C (<40 mg/dl)	8 (25.0%)	28 (100%)	9 (65%)	4 (5.1%)
Triglycerides (>200 mg/dl)	16 (50.0%)	19 (67.8%)	9 (65%)	13 (16.8%)

Table 3
Factors associated with Creatinine Clearance after adjustment.

	Category A: no nephropathy or microalbuminuria only; CCre≥90 ml/min/1.73m^2	Category B: mild to moderate reduced CCre, CCre: 30–89 ml/min/1.73m^2	Category C: severe reduced CCre or need for renal replacement treatment, CCre <30 ml/min/1.73m^2
N			
Male/Female	0.51	0.55	0.67
Age >40 years	0.47	0.49	0.54
Diabetes duration >10 yrs	0.43	0.47	0.79
Ulcer duration >1 month	0.69	0.73	0.12
Ulcer size >4 cm^2	1.74	1.51	1.34
Grade of ulcer (Wagner)	0.39	0.37	0.45
Amputation	0.57	0.51	0.67
Discharge Status	1.24	1.29	1.31
Alive	3.03	2.07	3.45
Died	2.05	2.17	2.42
Hospital stay (>1month)	5.16	5.87	5.56
HbA1c (>7%)	9.60	6.30	5.66
WBC count (10^3/μl)	3.03	2.43	3.45
Hb (g/dl)	2.48	2.84	2.56
Serum creatinine (>1.5 mg/dl)	2.84	3.23	3.67
SGOT/AST (>34IU/L)	2.32	2.57	2.49
SGPT/AST (>35 IU/L)	5.37	5.78	3.45
LDL-C (>100 mg/dl)	1.25	1.78	1.65
Total cholesterol (>150 mg/dl)	2.2	2.12	2.78
HDL-C (<40 mg/dl)	1.43	1.37	1.67
Triglycerides (>200 mg/dl)	1.62	1.26	2.67

Values are adjusted (Odds ratio) OR. Results represents adjustment for all variable listed in the tables.

BMI ($p = 0.040$), gender and BMI ($p = 0.031$), hospital stay and BMI ($p = 0.009$), HbA1c and BMI ($p = 0.031$), Hb and BMI ($p = 0.025$) and triglyceride and BMI ($p = 0.031$) (at 5% level of significance). In the context of HbA1c, findings have shown a positive association with gender ($p = 0.035$), diabetes duration ($p = 0.001$), ulcer duration ($p = 0.031$), amputation rate ($p = 0.020$), hospital stay ($p = 0.019$), Hb ($p = 0.015$), SGOT/AST ($p = 0.012$) and cholesterol level ($p = 0.018$). In the context of serum creatinine, findings have shown positive association with gender ($p = 0.029$), diabetes duration ($p = 0.029$), ulcer size ($p = 0.010$), grade of ulcer ($p = 0.014$), amputation rate ($p = 0.030$), hospital stay ($p = 0.005$), Hb ($p = 0.041$), SGOT/AST ($p = 0.042$) and triglyceride ($p = 0.031$).

Table 5 has shown the microorganism frequency of aerobic, anaerobic and fungal isolates from diabetic foot ulcers among foot ulcer patients. A total of 302 isolates were found, averaging of 1.28 per patients. The findings have shown that 204 aerobic isolates had polymicrobial pattern of growth while 51 aerobic isolates have monomicrobial type of growth. All isolates of anaerobic and fungal origin had polymicrobial aetiology. Furthermore, the findings have shown that Anaerobic isolates were present among patients with polymicrobial growth pattern (100%). Similarly, fungal isolates were present among patients with polymicrobial growth pattern (100%) as compared to monomicrobial growth pattern.

4. Discussion

The study has shown a better understanding towards the association between HbA1c and serum creatinine with microbiological profiling of infected ulcer cases. The findings have shown a positive correlation of HbA1c and serum creatinine with different microbiological parameters. These findings have been supported by previous studies. For instance, the results obtained by Crews et al. [8] showed that aerobic exercise has a significant impact on the patient with peripheral neuropathy and also reduced the

Table 4
Pearson Correlation analysis between BMI, HbA1c and Serum Creatinine with clinical and laboratory in patients with diabetic foot.

N	BMI	HbA1c	Serum Creatinine
	p value	p value	p value
Male/Female	0.031	0.035	0.029
Age >40 years	0.489	0.089	0.119
Diabetes duration >10 yrs	0.500	0.001	0.029
Ulcer duration >1 month	0.041	0.031	0.027
Ulcer size >4 cm^2	0.355	0.315	0.010
Grade of ulcer (Wagner)	0.485	0.415	0.014
Amputation	0.040	0.020	0.030
Discharge Status	0.368	0.218	0.128
Alive	0.365	0.215	0.123
Died	0.362	0.112	0.312
Hospital stay (>1month)	0.009	0.019	0.005
HbA1c (>7%)	0.031	0.011	0.021
WBC count (10^3/μl)	0.485	0.185	0.175
Hb (g/dl)	0.025	0.015	0.041
Serum creatinine (>1.5 mg/dl)	0.576	0.176	0.076
SGOT/AST (>34IU/L)	0.052	0.012	0.042
SGPT/AST (>35 IU/L)	0.248	0.108	0.048
LDL-C (>100 mg/dl)	0.489	0.119	0.089
Total cholesterol (>150 mg/dl)	0.068	0.018	0.068
HDL-C (<40 mg/dl)	0.129	0.119	0.029
Triglycerides (>200 mg/dl)	0.031	0.011	0.031

neuropathic symptoms in diabetic patients. Pain and depression are common among the patients of diabetes such that, musculoskeletal pain among these patients is approximately 1.7–2.1 more as compared to the patients without diabetes. Many diabetes patients suffer neuropathic pain, which can be reduced by aerobic exercise. Moreover, toe was found to be the most common ulceration point in body (54%) in the study conducted by Perim et al., [9]. The study also identified a total of 89 bacterial isolates among 30 patients. The results of the study showed that *Staphylococcus aureus* was found to be the most commonly isolated Gram-positive bacteria followed by other bacteria. *Staphylococcus* was also found in our study long with other aerobic microorganisms. Furthermore, several other microorganisms were also evaluated along with antibiotic susceptibility of isolates from the infected wound.

Table 5
Microorganism frequency of aerobic, anaerobic and fungal isolates from diabetic foot ulcers patients.

		N (%)	Growth pattern	
			Monomicrobial	Polymicrobial
AEROBIC		**255**	**51**	**204**
1	*Staphylococcus aureus*	*60*	8	52
2	*Enterococcus faecalis*	*9*	3	6
3	*Beta hemolytic streptococcus*	*6*	3	3
4	*CONS*	*6*	3	3
5	*Corynebacterium sp*	*7*	0	7
6	*Escherichia coli*	*71*	15	56
7	*Pseudomonas aeruginosa*	*40*	15	25
8	*Klebsiella oxytoca*	*18*	0	18
9	*Klebsiella pneumoniae*	*15*	2	13
10	*Proteus vulgaris*	*9*	0	9
11	*Proteus mirabilis*	*4*	0	4
12	*Acinetobacter sp*	*8*	2	6
13	*Morganella morganii*	*2*	0	2
ANAEROBIC		**21**		**21**
14	*Peptostreptococcus spp*	10		10
15	*Propionibacterium spp*	5		5
16	*Clostridium perfringens*	3		3
17	*Eggerthella lenta*	1		1
18	*Bacteroides ureolyticus*	2		2
FUNGAL		**26**	**-**	**26**
19	*Candida albicans*	16	–	16
20	*Candida dubliansis*	2	–	2
21	*Aspergillus sp*	6	–	6
Total		**302**	**51 (16.8)**	**251(83.1)**

Kumar et al. [10] linked vasculopathy, neuropathy and lower-extremity wound healing with the level of glycosylated haemoglobin. Maintaining normal levels of HbA1c had been shown to be very effective in the long-term management of glycemic levels of diabetic patients. Seventy percent of the patients suffered from foot trauma and neuropathy, and thirty percent of patients were suffering from vasculopathy. Moreover, the average healing time was 42 days. In the 12-week duration, patients with low levels of HbAlc showed faster healing. However, no improvements were seen in patients, suffering from vasculopathy or neuropathy [10].

A study was conducted to evaluate the creatinine levels in patients with diabetes and to examine its effect on Impaired Glucose Tolerance patients. Various demographic parameters like blood pressure, body mass index, waist hip ratio, socioeconomic status and age were taken into consideration. Additionally, Oral Glucose Tolerance Test (OGTT) too was carried out. A chemistry analyzer was used to measure uric acid, creatinine, bilirubin, HbA1c, lipid profile and urea levels of the patients. A strong association was found between HbA1c and serum creatinine. Age was a big factor that was observed in the increase of the creatinine level of IGT patients. No significant link was found between creatinine and low-density lipoprotein, TG and cholesterol [11].

Niveditha et al. [12] investigated the relationship between diabetic nephropathy and retinopathy with glycosylated hemoglobin. Serum creatinine, HbA1c, and urea levels tests of the patients were conducted. Out of the 50 patients, 8% of them were found to have HbA1c that was under very good control, where HbA1c was less than 6. Furthermore, 14% patients had HbA1c under good control, that translated as HbA1c between 6 and 8, and 78% patients were diagnosed with HbA1c that was under poor control (HbA1c more than 8). 26 patients had low serum creatine levels and 24 had high creatinine levels. Apart from this, 19 patients had high blood urea labels and 31 had low urea measurements. The relationship between HbA1c and serum creatinine showed that out of the 24 patients with high serum creatinine levels, 20 patients had glycosylated hemoglobin that was a very high value (HbA1c more than 8). Therefore, it was concluded that there was a positive relationship between serum creatinine levels and the HbA1c values. The risk of Diabetic nephropathy increases with an increase in both the creatinine and the HgA1c levels [12].

Diabetic foot ulceration is associated with high mortality and amputation rate. The amputation risk in the diabetic patients varies according to the severity of the disease. The amputation found in present study was 0.5% for category patients, 0.51% for category B patients, and 0.67 for category C patients. The study conducted by Wang et al. [13] showed that the frequency of amputations should be increased in the diabetic patients. According to Chinese Diabetes Society, the amputation rate was found to be 7.3% in the Chinese diabetic foot disease patients; however, 28.2% amputation rate was found to be caused by diabetes. Non-traumatic foot amputation is the most common among diabetic patients suffering from diabetic foot ulceration; thus, this can be reduced by incorporating multidisciplinary team for significant results.

Other factors which exacerbate the diabetic foot ulceration involves several fungal isolates, which includes *Candida albicans*, *Candida dubliansis* and *Aspergillus* sp as found in this study. Moreover, the study conducted by Kumar et al. [14] showed that the fungal aetiology was found in 75 patients out of 155 cases. All the fungal aetiology were identified as *Candida* species. The different *Candida* species present were *C. albicans*, *C. Krusei*, *C. parapsilosis*,

C. glabrata, and *C. tropicalis* [15,16]. Thus, the results of the study were found to be partially consistent with the present study.

5. Conclusion

The study has evaluated the association of HbA1c and serum creatinine levels among diabetic patients with foot ulcerations. The findings have indicated that WBC count, SGOT/AST (>34IU/L), and SGPT/AST (>35 IU/L) were positively and significantly associated with HbA1c, serum creatinine and BMI. Therefore, it has been concluded that neuropathy has a direct relationship with HbA1c. Future studies should consider these factors to assess the association of HbA1c and serum creatinine with microbiological profiling of infected ulcers.

Conflicts of interest

The author declares no conflict of interest.

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