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A Study on the Prevalence of Vitamin D Deficiency and Urinary Tract Infection in Type 2 Diabetes Patients at a Tertiary Care Centre in Tiruchirappalli

KEYWORDS: Diabetes mellitus, Vitamin-D, UTI

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ABSTRACT *Diabetes mellitus is the common endocrine disorder in developed and in developing countries like India. It has been hypothesized that insufficient Vitamin-D (25-hydroxy vitamin D) levels and urinary tract infections (UTIs) are most common and may contribute to pathogenesis of diabetes. The aim of the present study is to assess the prevalence of Vitamin-D deficiency and UTIs in type 2 diabetics. Serum Vitamin-D levels is analysed using ELISA kit and uropathogens (viz., E.coli, Enerobactericeae, Staphylococcus saprophyticus, and Enterococci) are isolated and identified from urine samples using standard micorbiological techniques. The results showed significant Vitamin-D deficiency and UTI occurrence to be highly prevalent in diabetic women than in men and the high prevalence in the age group of 35-55 years.*

INTRODUCTION

Diabetes mellitus (DM) is becoming a phenomenal worry to mankind in the new millennium, evident by dramatic increase in its prevalence and global incidence of about 387 million (8.3%). About one in 12 persons have diabetes and one out 2 don't know that they have diabetes. In 2014, about 4.9 million deaths have occurred due to diabetes (IDF, 2014). India is the second leading with 65.1 million diabetics following China with 98.4 million. According to IDF, this will increase upto 109 million by 2030. Besides other forms type 2 diabetes accounts for 85%-95% of all diabetes.

There is accumulating evidence to indicate the role of Vitamin-D deficiency in the development of type-2 diabetes. Observational studies suggest that Vitamin-D may lower the risk of type 2 diabetes. However, data from long-term trials are lacking. Animal studies describes Vitamin-D as a basic factor necessary for normal insulin secretion, thus reduces insulin resistance probably through its effect on calcium and phosphorus metabolism and through up regulation of the insulin receptor gene (Talaei et al., 2013). Vitamin-D has been associated with several of the contributing factors known to be linked to the development of type 2 diabetes (Danescu et al., 2009). Evidences indicate that circulating concentrations of Vitamin-D (25-hydroxy vitamin D) may be inversely related to the prevalence of diabetes, as both insulin resistance and impaired pancreatic beta cell function have been reported with Vitamin-D insufficiency. Moreover a Vitamin-D supplementation study shows beneficial influence in glycemic condition by optimizing glucose metabolism (Pittas et al., 2007).

Urinary tract infections are more common, more severe, and carry worse outcomes in patients with type 2 diabetes mellitus. Various impairments in the immune system, poor metabolic control, and incomplete bladder emptying due to autonomic neuropathy may all contribute to the enhanced risk of urinary tract infections in these patients (Nitzan et al., 2015). Although the exact mechanism is unknown, there are several possible explanations for the association between diabetes and urinary tract infections (UTIs). One theory is that damage to the genitourinary system as a result of diabetic neuropathy could lead to a dysfunctional bladder creating more opportunity for infection. The presence of higher glucose concentrations in the urine may present another possible pathway by allowing for increased bacterial reproduction and a favorable environment for infections. Last, impaired immune response may play a role in a patient's decreased ability to fight off bacteria (Fu et al., 2014).

The aim of the present study is to assess the prevalence of Vitamin-D deficiency and uropathogens causing UTIs in type 2 diabetes patients at a tertiary care centre in Tiruchirappalli district, Tamil Nadu.

MATERIALS AND METHODS

Study area and participants

The present study was conducted in a tertiary health care centre in Tiruchirappalli, Tamil nadu. Men and women were eligible to participate in the study if they had uncontrolled type 2 diabetes mellitus, with fasting plasma glucose > 140 mg/dl and glycosylated hemoglobin > 7.0 %. 300 diagnosed cases of type 2 diabetes belonging to age group of 35-75 years were included in this study. 300 age matched subjects who have no history of diabetes mellitus, UTI and Vitamin-D deficiency were taken as controls. All were enrolled in the study after signing the informed consent form.

Assessment of Vitamin-D

Blood samples (5ml) were collected by venipuncture and sera separated by centrifugation (3000rpm/5min). 25-hydroxy Vitamin-D (25OHD) levels were estimated by using Enzyme linked immunosorbent assay (ELISA) kit. All the results were expressed as mean ± SD and statistical comparisons were done using student t-test using the SPSS package. The criteria used to define Vitamin-D status were: sufficiency (25OHD levels, 30-100 ng/mL), insufficiency (25OHD, 20 to 30 ng/mL), and deficiency (25OHD, < 20 ng/mL).

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Prevalence of UTI

Midstream urine samples (10-20ml) were collected using sterile wide-mouth universal containers and stored at 2-8^{0}C in the refrigerator until culture time. Urine specimen were directly inoculated on to blood agar, Cysteine Lactose Electrolyte Deficient (CLED) media, and MacConkey agar (Oxoid Ltd, Basing stroke, Hamsphire, England) using a standard calibrated wire loop (0.002ml) streaked culture plates were incubated at 37^{0}C over night. On the next day, the bacterial growth on the respective media was observed, and total colony count was done on blood agar and checked for significant bacteriuria.

Plates were observed under the microscope for bacterial growth after 24-48hours. Colonies greater than 30 or 104 colony forming units per millimeters were considered significant. Isolated colonies after purification were initially Gram stained. By using Bergey's Manual of determinative bacteriology, the isolates were biochemically (*viz.*, coagulase test, oxidase test, indole) characterized and identified. All the bacterial isolates were preserved on nutrient agar slants at 4^{0}C and sub-cultured periodically for further studies.

RESULTS AND DISCUSSION

Diabetes mellitus comprises a group of common metabolic disorders that share the phenotype of hyperglycemia. The greatest number of people with diabetes is between 40 and 59 years of age (IDF, 2014). Vitamin-D deficiency/insufficiency, associated with increased insulin resistance, systemic inflammation and HbA1c in type 2 diabetics, is now regarded as pandemic in all age groups as has been documented previously (Goswami et al., 2000; Cigolini et al., 2006) Pittas et al., (2007) demonstrated that pancreatic insulin secretion is inhibited by Vitamin- D deficiency. The mechanisms by which Vitamin-D may affect the risk of type 2 diabetes are not clear. Studies on associations between insulin secretion and Vitamin-D have been inconsistent. The results of serum Vitamin-D analysis is depicted in table 1.

Table 1: Prevalence of Vitamin-D deficiency

Age group	Sex group	No. of cases with Vitamin D deficiency (<20ng/ml)	
		Control	Diabetics
35-45	Male (n=25)	-	8
	Female (n=25)	8	12
45-55	Male (n=50)	24	36
	Female (n=50)	28	48
55-65	Male (n=50)	19	33
	Female(n=50)	26	46
65-75	Male (n=25)	6	16
	Female (n=25)	12	18
Total	300	123	217

There have also been many suggestions about the association of Vitamin-D deficiency and the incidence of type 2 diabetes mellitus. Studies have suggested an inverse relationship between low intake of total Vitamin-D and risk of type 2 diabetes (Pittas et al., 2006). In a recent study, Kedari et al., (2014) reported significant decrease in the levels of Vitamin-D in type-2 diabetics. In our study, significant decrease in Vitamin-D level was obsereved in about 41% of healthy controls and about 72.3% of diabetics and was found to be deficient. It has already been reported that the prevalence of Vitamin-D deficiency in women with diabetes is more common than in men with diabetes (Isaia et al., 2001). This study reveals that the prevalence of Vitamin-D deficiency in both healthy controls and diabetic women (49.6%) is more than in men (31%). It has also been observed that the prevalence of Vitamin-D deficiency if higher in the age group of 45-65 years in both healthy (58.5%) and diabetic (81.5%) study population.

UTI is a common infection observed in diabetic patients. Escherichia coli are the most common bacterial pathogen causing urinary infection in diabetics, other organisms being Klebsiella pneumoniae, Proteus mirabilis and Pseudomonas aeroginosa. Fu et al., (2014) reported common baseline occurrence as well as recurrence of UTI in type 2 diabetics than non diabetics. According to IDF (IDF, 2014), there is about 54% of increased risk of UTI in type 2 diabetics than in non diabetics. The prevalence of uropathogens responsible for UTI in type 2 diabetics is represented in table 2.

Table 2: Prevalence of uropathogens responsible for UTI in type 2 diabetes

Uropathogens	Urinary Tract Infection	
	Control	Diabetics
Eschericia coli	28%	52%
Klebsiella spp.	-	24.4%
Enterococcus spp	1.06%	7.62%
Staphyloccus aureus	3.2%	7.48%
Enterobacter spp	-	4.82%,
Pseudomonas aeruginosa	-	2.07%
Proteus spp	-	1.61%

Demonstrative research studies have substantiated the significance of diabetes in relation to UTI and sufficient evidence in the form of data has been provided to imply the role of diabetes in UTI. In our study it has been observed that the overall prevalence of UTI was 26% in non diabetics, 57% in diabetics. The most prevalent age group is the 35-55 years. In diabetics UTI was observed 54% of females and 42% of males. Significant bacteriuria was seen in 48%, 32% had insignificant colony count and no growth was seen in 20% of the specimens. About 162 uropathogens (128 gram negative (79.01%) and 34 gram positive (20.9%) were isolated from the diabetic population. Out of them, E. coli (52%) was the most common, followed by Klebsiella spp. (24.4%), Enterococcus spp (7.62%), Staphyloccus aureus (7.48%) while Enterobacter spp (4.82%), Pseudomonas aeruginosa (2.07%) and Proteus spp (1.61%). This result is similar to that of previous work carried out by Bashir et al., (2008) in which the isolates were Eschericia coli (64.3%), followed by Staphylococcus aureus (21.4%), and Klebsiella pneumoniae (14.3%). Studies to validate the fact of growing resistance among UTI causing pathogens have been going on for the last three decades and the available data and reports confirm that the increase in resistance to commonly employed antibiotics is a consequence of inappropriate use of the antimicrobial agent.

CONCLUSION

In summary, type 2 diabetics have worse outcomes than non diabetics. Vitamin-D deficiency occurs in both groups with higher prevalence in women than in men. The age group with worse outcomes of vitamin-d is between 45 and 65. The prevalence of UTI was high in diabetic wom-

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en than in diabetic men with E. coli as the most common uropathogen. Since type 2 diabetes is global burden treatment with Vitamin-D supplementation and appropriate antibiotics with higher efficiency should be suggested to subside the worst overcomes suffered by diabetics.

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