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SERUM B_{12} STATUS AMONGST TYPE 2 DIABETES MELLITUS ADULTS ON METFORMIN

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ABSTRACT

Of adults on metformin, 5.8–33% has evidence of reduced vitamin B12 absorption. No guidelines of standard care for B12 screening and treating low B12 levels in Type 2 Diabetes Mellitus (T2DM) adults exists. B12 malabsorption by metformin can result in B12 deficiency due to calcium-dependent ileal membrane antagonism, potentially leading to neuropathy microvascular complication deteriorating the quality of life in T2DM adults. The objective of this study was to determine the prevalence of B12deficiency in T2DM adults and to understand its implications in management of T2DM. 144 metformin treated T2DM adults were assessed for nutritional status and biophysical measurements. Of 144 patients on 75, vitamin B_{12}, Hb1Ac and Hemoglobin with cell morphology were done using CLIA , HPLC and C.B.C respectively. A case control design was employed using two groups (B12<200pg/dl and B12>200pg/dl) to find association of B12 and other factors**.** Vitamin B_{12} deficiency (<200pg/ml) was recorded in one third (36%) of the adults between 30-96 yrs. B12 deficiency was more among females(77%)**;** majority (30%) with very low (<150pg/ml) B12 levels. None showed macrocytic anemia. More than one-fourth(38%) reported anorexia and metallic taste as metformin side effects ($p<0.05$).Adults on higher dosage showed greater prevalence(75% vs 30%) of metallic taste. Of 27 B12 deficient adults only 5(18%) had euglycemia (Hb1Ac<7%); fundamental to prevent microvascular and neuropathic complications. Odds of glycation and gastrointestinal side effects of metformin were 4.05 and 1.43 respectively. B12 deficient diabetics are more likely to have poor glycemic control and gastrointestinal side effects of metformin therapy. They are prone to anorexia and metallic taste, potentially leading to difficulties for dietary compliance; a corner stone in improving glycemic control and thereby reducing the risk of neuropathies. B12 deficiency seems crucial as regards metformin side effects and glycemic control.

Key Words: Vitamin B12 deficiency; metformin side effects; glycemic control, type 2 diabetes

INTRODUCTION

Vitamin B12 deficiency is known to be common in vegans which can give rise to hematological and neurological manifestations. Metformin treated type 2 adults (T2DM) are prone to develop low serum vitamin B12 levels due to vitamin B12 malabsorption by this essential antidiabetic medicine (American Diabetes Association, 2009 and WHO, 2009) . Metformin use in T2DM adults has also been shown to cause B12 deficiency in some studies (Reinstatler et.al., 2012, De et.al., 2010, Liu et.al., 2011, Wile, 2010 and Dustan et.al., 2012). Some authors have also questioned the clinical relevance of these findings since B12 deficiency is asymptomatic at most times (Pflipsen et.al., 2009). Some case reports have emerged showing that metformin-induced B12 deficiency can cause neuropathy, which may be attributed to poor glycemic control in type 2 diabetes patients and may be left untreated. Till date no guidelines to recommend B12 deficiency cut offs in diabetes has been recommended by ADA or IDF.

Decrease in vitamin B12 absorption and levels following metformin use typically starts at fourth month. Recent studies determine prevalence of metformin-induced B12 deficiency (<200pg/ml) ranging from 5.8% to 33% (8, 11, 12). This wide range is due to a varied study definition of B12 deficiency. In India serum B12 levels have been studied in free living population by Yagnik et.al[16]. B12 levels have been reported in T1DM in South India among 90 patients(45.5% by the cutoff point of <180 pg/ml and 54% using the published cut off point of <200 pg/ml)[15]. Recently, Singh and coworkers[13] showed that proportion of patients with B12 deficiency (150-220 pg/ml) in metformin exposed group was significantly higher than proportion in non metformin exposed group (21.4% vs.5.7%).

Due to paucity of Indian studies evaluating the prevalence of B12 deficiency in T2DM adults on metformin, we planned a cross sectional study employing a case control design to compare B12 deficient to those with normal B12 status among T2DM adults on metformin.

MATERIAL AND METHODS

SETTING AND PARTICIPANTS

In a period of six months (December 2013-May 2014) 144 T2DM adults attending a family medicine clinic were enrolled in study only if they were on metformin therapy for a minimum of 4 months duration. Participants with high creatinine levels (1.7 mg/dl for men and 1.5 mg/dl) for women, hypothyroidism and prescription B12 injections and proton pump inhibitors were excluded from the analysis. We also excluded pregnant T2DM women and those without diabetes taking metformin. Based on clinical aspects described by the American Diabetes Association participants who reported receiving a physician's diagnosis after age 30 (excluding gestational diabetes) and did not initiate insulin therapy within 1 year of diagnosis were classified as having type 2 diabetes. They were screened for nutritional status and biophysical measurements. Of 144 patients on 75, vitamin B_{12}, Hb1Ac and Hemoglobin with cell morphology were done using CLIA, HPLC and C.B.C respectively.

OUTCOMES AND FOLLOW-UP

The primary outcome was biochemical B12 deficiency determined by serum B12 concentrations. Serum B12 levels were quantified using vitamin B12 radioassay kit from Sir Ganga Ram hospital laboratories, New Delhi. We defined biochemical B12 deficiency at serum levels < 200pg/ml, very low as serum B12 between 149 to 200 pg/ml, and normal as above 200 pg/ml. Using data collected in the B12 screening proforma study participants were classified as currently using metformin therapy (alone or in combination therapy) versus those not presently using metformin. In the final analysis, case control design was employed where two groups were used to allow the comparison of those diagnosed as B12 deficiency (<200pg/ml) versus those with normal B12 status (>200pg/ml) among T2DM on metformin. To determine the association between metformin and biochemical B12 deficiency data on the metformin side effects and other factors were used.

STATISTICAL ANALYSIS

Statistical analysis was done by using SPSS version 16.0 and Epi Info version 7. Means, range, frequencies and percentages were calculated. Independent t test and χ^2 tests were employed as test of significance to infer the results from sample to population. Odds ratio and Pearson correlations were applied to the data to find associations.

RESULTS AND DISCUSSION

RESULTS

Most of the demographic and biological characteristics (Table 1) were similar in both genders with no significant difference between the two, except for years of diabetes being significant ($p<0.001$). Among T2DM adults on metformin, the mean age was 56.98±0.98years, with 34.7% being males. A median duration of disease was 5 years. On an average the T2DM adults were overweight (Mean BMI 26.74± 4.73) and Pre hypertensive (Mean SBP/DBP 129.4/81.70) as categorized by Asia Pacific Classification by WHO 2000[17] and 8th JNC Report[18] respectively (Table1). Blood pressure status when related with B12 levels revealed significant associations. Compared with those with normal B12 levels (>200), B12 deficient (<200pg/ml) adults had greater proportion of pre hypertensive adults (64.6% vs 70.4%) and there was a significant relationship at 5% level between Hypertension and B12 status of adults on metformin ($\chi2 = 12.68$, d.f=3,p=0.005). A gender wise comparison of the biochemical profile of T2DM adults is given in Table 2.

Table 1: Demographic and biological characteristics of Indian T2DM adults by gender

	Males (N=50)	Females (N=144)	Test Statistic	P value
Age,Mean(SE)	58.00(1.93)	56.30(1.14)	0.81	$0.42^{N.S}$
Years of Diabetes,Mean(SE)	6.51(0.53)	6.33(0.65)	7.32	0.000**
Months on Motorman	4.26(0.17)	4.52(0.11)	1.38	$0.17^{N.S}$
BMI, Mean(SE)	26.64(4.82)	26.79(0.49)	0.17	$0.86^{N.S}$
W.C,Mean(SE)	90.24(1.45)	90.63(1.30)	0.19	$0.85^{N.S}$
H.C,Mean(SE)	93.62(1.52)	96.48(2.10)	0.93	$0.36^{N.S}$
S.B.P Mean(SE)				
D.B.P Mean(SE)				
BMI(Kg/m$^{2)a}$,n(%)			X^2	
(Uw)Underweight(<18.5)	2(50)	2(50)	3.99	$0.40^{N.S}$
(N)Normal(18.5-22.9)	8(38.1)	13(61.9)		
(Ow)Overweight(23-24.9)	11(33.3)	22(66.7)		
(Ob)Obese(25-29.9)	15(26.8)	41(73.2)		
(Mor.Ob)Morbid Obese(>30)	14(46.7)	16(53.3)		
B.P(S.B.P/D.B.P)^{b} , n(%)				
Normal	3(6)	1(1.1)	3.14	$0.37^{N.S}$

Prehypertensive	31(62)	61(64.9)		
Stage I Hypertension	12(24)	22(23.4)		
Stage II Hypertension	4(8)	10(10.6)		

a=W.H.O 2000[17],b=J.N.C VII 2007[18],c=WHO 1986[19]

Table 2: Biochemical profile and distribution of the T2DM adults on metformin based on several biochemical deficiencies

	N	Males	N	Females	t value	P value
Hemoglobin(g/dl),Mean(SE)	39	7.46(0.9)	81	8.09(0.63)	0.56	$0.57^{N.S}$
Serum B_{12}(pg/ml),Mean(SE)	24	186.29(22.6)	55	227.68(22.39)	1.11	$0.27^{N.S}$
Hb1Ac(%),Mean(SE)	50	7.8(0.32)	94	7.70(0.28)	0.21	$0.83^{N.S}$
					X^2	
Anemia$ n(%)	25	20(80)	56	26(46.4)	7.84	0.005**
Normal Hb, n(%)	25	5(20)	56	30(53.6)		
B_{12} deficiency(pg/ml)	75					
Normal B12 (>200pg/ml)	48	16(33.3)		32(66.6)	1.01	$0.31^{N.S}$
B_{12} Deficient(<200pg/ml)	27	6(22.3)		21(77.7)		
Glycemic Control# (%Hb1Ac)	135					
Good	45	17(37.7)		28(62.2)	0.144	$0.70^{N.S}$
Poor	90	31(34.4)		59 (65.5)		

$Hemoglobin<13g/dl for males, <12g/dl for females (WHO 1986)[20], #<7%Good,>7%Poor *Significant at 5%

Table 3: Glycemic control Compared with B12 status deficiency status

Serum B12 (pg/ml)	**N=75**			
	T	**Good Hb1Ac (6.00-7.49)%**	**Fair Hb1Ac (7.50-8.99)%**	**Poor Hb1Ac (9.00-15.00)%**
		n (%)	n(%)	n(%)
Normal B_{12} (>200)	48	25(53.2)	14(29.8)	8(17.0)
B_{12}Deficient(<200)	27	7(25.9)	9(33.3)	11(40.7)
X^2	6.75			
P value	0.034*			

*Significant at 5%

Table 4: B12 Status associated with Metformin Dosage and GI Side Effects of Metformin

Serum B12 (pg/ml)	**GI Side Effects of Metformin**			
	T	**No Side Effects**	**Anorexia**	**Metallic Taste**
	75			
Normal B_{12}(>200)	48	28(58.3)	10(20.8)	10(20.8)
B_{12} Deficient(<200)	27	18(66.7)	0(0)	9(33.3)
X^2			6.88*	
Present Metformin Dosage (mg/day)	144			
Metformin GI Side Effects	36	46 25(69.4)	28(60.9) 4(11.1) 1.43	7(19.4) 0.53-3.82
Values in parenthesis are in percent; p<0.05* More than half (56%) of the adults had border line low B12(151-400pg/ml) as compared to a small proportion **500**				
>500≤1000	81	45(55.6)	12(14.8)	24(29.6)
≥1500-2500	27	7(25.9)	4(14.8)	16(59.3)
X^2	13.88*			
Serum B12 (pg/ml)				
	Past Metformin Dosage (N.R=2)			
		500 mg/day	>500-1000 mg/day	≥ 2000 mg/day
Normal B_{12}(>200)	48	17(35.4)	25(52.1)	4(8.3)

B12 Deficient(<200)	27	19(70.4)	7(25.9)	1(3.7)
X^2	8.85*			

Values in parenthesis are in percent; p<0.05*,N.R=No Response

Table 5 Association of several factors with B12 levels of T2DM adults on metformin

		Serum B_{12} levels (N=75)		O.R	95% CI
Factors	**N**	**<200pg/ml**	**>200pg/ml**		
Hypertension (SBP>120/DBP>80)	73	25(34.2)	48(65.7)	-	-
WC (M≥90, F≥ 80)	57	23(40.3)	34(59.6)	2.37	0.69-8.10
BMI(>23 Kg/m^2)	63	23(36.5)	40(63.4)	1.15	0.31-4.24
WHR>1	15	3(20)	12(80)	0.37	0.09-1.47
Glycated Hb(>7%)	47	22(46.8)	25(53.1)	4.05*	1.31-12.4
Duration T2DM(>10yrs)	10	4(40)	6(60)	1.22	0.31-4.7
Metformin Mono Therapy	31	13(41.9)	18(58.1)	1.55	0.60-4.02

(12%) of the adults with optimal levels (>400pg/ml). B12 deficiency (<200pg/ml) was more common in females than males (77.7%vs.22.3%) however significantly greater proportion of males than females (80%vs.20%, p<0.05) were anemic. This could be attributed to the practice of prescribing iron supplements more commonly to females than males (20%vs.4%). More Females than males (62.2%vs.37.7%) showed good glycemic control. This may be due to the fact that females being more conscious of their body image visit dietitian more commonly than males; contact with dietitian making females more compliant to dietary advice, crucial for glycemic control beyond their compliance to oral hypoglycemic agents. Cell morphology reported no case of macrocytic anemia in our study where a minimum of 94 pg/ml B12 levels were seen.

Serum B12 levels when associated with the Hb1Ac (Table 3) showed that cases with low B12 (<200pg/ml) had significantly greater proportion of poorly controlled diabetes (14.7% vs.17%) as compared to the controls with normal B12 status (>200pg/ml).Significantly greater proportion of people with good glycemic control had normal B12 levels compared to those with low B12 levels.

There was a significant relationship at 5% significance level between the dosage of metformin (last reported in prescription) and their recent B12 status (Table 4). Also a significant relationship at 5% significance level between gastrointestinal (GI) side effects of metformin with their present metformin dose and their B12 status. One third (33.3%) of B12 deficient (<200pg/ml)T2DM adults reported metallic taste to be the most common side effects of metformin; detrimental to their food ingestion required for dietary compliance; crucial for maintaining euglycemia in order to prevent micro vascular secondary complications and neuropathy and thereby maintaining their quality of life.

Among the several factors associated with B12 deficiency by odds (Table 5) the abdominal obesity (WC),BMI, glycemic control, metformin mono therapy and GI side effects of metformin were associated (O.R>1). However glycemic control emerged as the only significant predictor for B12 deficiency on metformin treated T2DM adults. The odds of uncontrolled diabetes (Hb1AC>7%) was 4.05 i.e. a patient who has low B12 levels (<200pg/ml) is almost four times more likely to have uncontrolled diabetes than a patient who have normal B12 levels (>200pg/ml). The similar was reported by Nazni and Ravinder Singh (2014)

CONCLUSION

Our data demonstrates clear association between metformin and biochemical B12 deficiency among T2DM adults. Prevalence of B12 deficiency being more than 1% among T2DM adults can be stated as a problem of Public Health concern. There is paucity of literature on Indian population to state the B12 status of T2DM adults. Our results suggest several findings that add to the complexity and importance of B_{12} research and its relation to glycemic control in diabetes. It poses a need to study the relationship of B12 deficiency with diabetic neuropathy. Further the efficacy trials for B_{12} supplementation on national representative data should be done so as to draw guidelines to treat B_{12} deficiency among T2DM adults. Also the sustainability of B12 supplementation in relation to diabetic neuropathy needs to be established.

This study is subject to limitations. First, it is a cross sectional survey and it cannot assess time as a factor and therefore the results are only associations and not casual relationship. The cause and effect relationship between B12 deficiency and B12 supplementation among T2DM adults is recommended. A second limitation arises in our definition of biochemical B12 deficiency. There is no general consensus on how to define normal versus low B12 levels. Thirdly, owing to a small sample size the sample is not subjected to regression analysis and the association of B12 deficiency in relation to the dietary factors needs to be further studied.

Given that a significant proportion of the population of B12 deficient are more likely to have poor glycemic control and gastrointestinal side effects of metformin therapy making them prone to feeding problems like anorexia and metallic taste, potentially leading to difficulties for dietary compliance; a corner stone is to improve glycemic control thereby reducing the risk of neuropathies.

Addressing B12 deficiency seems crucial as regards metformin side effects and glycemic control to further prevent deterioration in quality of life due to neuropathy. Research exploring B12 in relation to diabetic neuropathy is scanty, especially peripheral neuropathy assessment in Indian T2DM adults is studied by few, however none had a national representative sample.

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ETHICS

All participants gave written in-formed consent, and the study protocol was approved by a Institutional Ethics Committee of the Maharaja Sayajirao University of Baroda.

REFERENCES

- American Diabetes Association. Standard of Medical care in diabetes-2009. Diabetes Care. 2009; 32:S13–61. [PMCID: PMC2613589] [PubMed: 19118286]
- WHO; 2009. World Health Organization.WHO Model List of Essential Medicines; p. 24. Available from : http://www.who.int/medicines
- Reinstatler L, Qi YP, Williamson RS, Garn JV, Oakley GP Jr. Association of biochemical B12 deficiency with metformin therapy and vitamin B12 supplements: The national health and nutrition examination survey, 1999- 2006. Diabetes Care 2012;35:327-33. [PUBMED]
- De Jager J, Kooy A, Lehert P, Wulffelé MG, van der Kolk J, Bets D, et al. Long term treatment with metformin in patients with type 2 diabetes and risk of vitamin B-12 deficiency: Randomised placebo controlled trial. BMJ 2010; 340:2181-7.
- Liu KW, Dai DL, Ho W Lau E, Woo J. Metformin-associated vitamin B12 deficiency in the elderly. Asian J Gerontol Geriatr 2011; 6:82-7.
- Wile DJ, Cory Toth. Association of metformin, elevated homocysteine, and methylmalonic acid levels and clinically worsened diabetic peripheral neuropathy. Diabetes Care 2010; 33:156-61.
- Dunstan DF, Rees JA, Chen S, Lansdown JA, Moat SJ, Ellis R, et al. An observational study of the effect of metformin on B12 status and peripheral neuropathy. Br J Diabetes Vascular Dis 2012;12:189-93.
- Pflipsen M, Oh R, Saguil A, Seehusen D, Topolski R: The Prevalence of Vitamin B12 Deficiency in Patients with Type 2 Diabetes: A Cross- Sectional Study. J Am Board Fam Med 2009, 22:528–34.
- Andres E, Serraj K.Optimal mgt. of pernicious anemia. Journal of blood medicine 2012,3:97-103.
- Hermann L, Nilsson B, Wettre S: Vitamin B12 status of patients treated with metformin: a cross-sectional cohort study. British Journal of Diabetes & Vascular Disease 2004, 4:401
- Reinstatler L, Qi Y, Williamson R, Garn J, Oakley-Jr G: Association of Biochemical B12 Deficiency With Metformin Therapy and Vitamin B12 Supplements. The National Health and Nutrition Examination Survey, 1999–2006. Diabetes Care 2012, 35:327–33.
- Qureshi S, Ainsworth A, Winocour P: Metformin therapy and assessment for vitamin B12 deficiency: is it necessary? Practical Diabetes 2011, 28:302–4.
- AK Singh, A Kumar, D Karmakar, RK Jha. Associations of B12 deficiency and clinical neuropathy with metformin use in type 2 diabetes patients. Journal of Postgraduate Medicine 2013, 59:4 253- 257:
- Nazni,P and Ravinder Singh, Meta analysis study of glycemic index of various food groups, International Journal of Food And Nutritional Sciences, 2014, Vol 3 Iss 4; pp-179-183.
- Adam JF, ClarkJS, IrelandJT, KessonCM, WatsonWS. Malabsorption of vitamin B12 and intrinsic factor secretion during biguanide therapy. Diabetologia 1983; 24:16-8.
- Kumthekar AA, Gidwani HV, Kumthekar AB. Metformin associated B12 deficiency. J Assoc Physicians India. 2012 Mar; 60:58-60.
- Yajnik C, Deshpande S, Lubree H, et al: Vitamin B12 deficiency, Hyperhomocystenemia in Rural and Urban Indians.
- Erdembileg Annurad, Kuninori Shiwaku, Akiko Nogi, Keiko Kitajima, Byambaa Enkhmaa, Kumiko Shimono and Yosuke Yamane The new BMI criteria for Asians by the regional office for the western pacific region of WHO are suitable for screening of overweight to prevent metabolic syndrome in elder Japanese workers, 2003, J Occup Health; 45: 335-343
- 2014 Evidence-Based Guideline for the Management of High Blood Pressure in Adults Report From the Panel Members Appointed to the Eighth Joint National Committee (JNC 8)
- WHO (1968). Techn. Rep. Ser. No. 405.