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308 Lasani Town, Sargodha Road, Faisalabad - Pakistan
Mob: +92 300 3008585, Fax: +92 41 8815544
E-mail: editorpjn@gmail.com

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Evaluation of Vitamin B_6 Status of Adult Saudis in Al-Qassim Region-Saudi Arabia

Khalid S. Al-Numair
Department of Food Sciences and Nutrition, College of Food and Agricultural Sciences,
King Saud University, P.O. Box 2460, Riyadh 11451, Saudi Arabia

Abstract: The aim of this study was to evaluate vitamin B_6 status of adult Saudis in Al-Qassim region-Saudi Arabia. A sample of 239 (127 males and 112 females) healthy Saudi adults was recruited. Vitamin B_6 status was evaluated by dietary intake and biochemical measurements. Results of dietary intake measurements showed that means of vitamin B_6 intake and intake of vitamin B_6 to protein ratio for males and females were 1.97±0.58 mg/day, 0.021±0.003 and 1.89±0.61 mg/day, 0.020±0.04, respectively. Results of biochemical measurements showed that means of plasma pyridoxal phosphate (PLP) concentration and average urinary 4-Pyridoxic Acid (4-PA) excretion for males and females were 29.6±9.6 nmol L^{-1} and 7.0±2.4 μmol day^{-1} and 31.7±10.4 nmol L^{-1} and 6.8±2.5 μmol day^{-1}, respectively. The study also investigated and discussed other health indicators that are related to vitamin B_6 status including Body Mass Index (BMI), plasma albumin concentration and alkaline phosphatase activity. Results of dietary intakes and biochemical measurements indicated an adequate status of vitamin B_6. There were no significant differences between males and females in any of the previous measurements. This study indicated an adequate status of vitamin B_6 among healthy adult Saudis in Al-Qassim region, however due to the numerous functions and the importance of the vitamin, the study suggested a further evaluation in various groups of the Saudi population in other regions of the country.

Key words: Vitamin B_6 intake, protein intakes, plasma PLP, red blood cell PLP, urinary 4-PA, plasma albumin, plasma alkaline phosphatase, adult Saudi

INTRODUCTION

The term vitamin B_6 refers to three primary forms of 3-hydroxy-5-hydroxymethyl-2-methylpyridine and their phosphorylated forms. The primary forms are Pyridoxal (PL), Pyridoxine (PN) and Pyridoxamine (PM). Their phosphorylated forms are PLP, PNP and PMP (Vasilaki *et al*., 2008). An additional form of vitamin B_6 that is found only in plant foods is glycosylated pyridoxine. Bioavailability of vitamin B_6 from animal food is quite high and could reach 100% (Reynolds, 1988); whereas the bioavailability of the vitamin from plant foods is lower compared to animal foods due to the presence of glycosylated pyridoxine as well as dietary fiber (Hansen *et al*., 1996). The phosphorylated forms of the vitamin must be dephosphorylated in the intestine by alkaline phosphotase prior to absorption. The forms of the vitamin are converted to their phosphorylated forms in the human liver and then to the physiological active form, Pyridoxal-5-Phosphate (PLP), which is released to blood bounded to albumin (Leklem, 1999). PLP participates as a coenzyme in more than 100 enzymatic reactions. The majority of these reactions are involved in amino acid metabolism (Vasilaki *et al*., 2008; Leklem, 2001). Thus, the requirement for vitamin B_6 increases with an increase intake of protein. A ratio of >0.016 mg vitamin B_6/g protein is required for adequate status of the vitamin (Hansen *et al*., 1997). Several studies have reported the high consumption of protein by Saudis. Al-Shoshan (1992) and Miladi (1998) reported a high consumption of red meat and poultry by Saudis. Al-Assaf and Al-Numair (2007) reported high consumption of protein by urban and rural adult Saudi males in Riyadh region. The study illustrated that although the intakes of protein were within the Acceptable Macronutrient Distribution Ranges (AMDRs) of Dietary Reference Intakes (DRI, 2005), they were high due to the high intakes of calories. Two studies have investigated protein and vitamin B_6 content of fourteen popular meat-based Saudi dishes (Sawaya *et al*., 1986) and six popular cereals and legume-based Saudi dishes (Al-Jebrin *et al*., 1985). The two studies found that vitamin B_6 content, compared to protein, was relatively low. Since high protein consumption among Saudis has been reported and based on the last two mentioned studies, an inadequate status of vitamin B_6 might be expected among Saudi population. Until present, there is no available study that has investigated the status of vitamin B_6 in any group of the Saudi population. Thus, the aim of this study was to evaluate vitamin B_6 status among adult Saudi males and females in Al-Qassim region by dietary and biochemical measurements. The dietary measurements included dietary intake of vitamin B_6, protein, vitamin B_6: protein ratio and % of vitamin B_6 from animal sources. The biochemical measurements included PLP concentrations in plasma and Red Blood Cells (RBC) and urinary 4-Pyridoxic Acid (4-PA). In addition, the study

evaluated two biochemical factors that affect plasma PLP, which are concentration of plasma albumin and plasma alkaline phosphotase activity.

MATERIALS AND METHODS

Recruitment of the sample from Al-Qassim region was done by personal contact and by advertisement. All volunteers received a questionnaire asked for age, weight, height, health status, exercise habits, use of medications and use of dietary supplements. For female volunteers, the questionnaire also asked for pregnancy, lactation and use of oral contraceptives. Users of dietary supplements or drugs that are known to affect vitamin B_6 metabolism, absorption or urinary excretion (Bhagavan, 1985) were excluded. Only healthy and non-users of medications or dietary supplements (non-pregnant, none lactating and non-users of oral contraceptives for females) were eligible to participate in the study. The study protocol was explained to each participant individually and informed consent was obtained. All participants received written instructions about keeping three consecutive days of food records and urine collection. For each participant, energy requirement (based on weight, height, age and physical activity level) and energy intake (based on his/her food record intake), were determined by the Food Processor (2001). Participant (s) for whom their difference between energy requirement and energy intake exceeded 15% was (were) excluded since this difference indicated incomplete or invalid food records. The study was carried out in 239 healthy Saudi adults, 127 males and 112 females and was performed between May and October 2008. The three consecutive day food records were analyzed by the Food Processor, version 7.8 (2001) (ESHA Research, Salem, OR, USA) to determine the dietary measurements. On the fourth day weight and height were measured and BMI was calculated for each participant. 15 ml of blood sample was collected from each participant in heparinized tubes (Becton Dickins, NJ, USA) by trained personal. Blood samples were centrifuged at 1800×g at 5°C for 20 min. and plasma samples were separated and stored at -70 (±5°C) until analysis. Red blood cells were washed three times using cold 0.9% saline and centrifugation of 1800×g for 20 min at room temperature (Miale, 1977). Red blood cells samples were stored at -70 (±5°C) until analysis. The urine samples were collected in brown opaque three-liter polyethylene bottles containing 15 ml of toluene as a preservative. Total volume of each day was measured and a sample of 10 ml was obtained and stored at -25°C until analysis. Plasma and Red Blood Cell (RBC) PLP were analyzed by HPLC with fluorometric detection as described by Kimura *et al.* (1996). Urinary 4-PA was analyzed by HPLC method as described by Gregory and Kirk (1979). Plasma albumin concentration was determined by colorimetric method as describe by Slater *et al.* (1975). Plasma alkaline phosphatase activity was measured by colorimetric method as described by Roy (1970). All samples were analyzed in duplicate and the average was reported if the difference did not exceed 8%.

Data analysis: Data analysis was performed using the Statistical Package for the Social Sciences, version 11.0 (SPSS) computer software. Descriptive statistics were adapted to display data in means ± SD and percentages. The statistical method of t-test was used to compare the mean values obtained between the males and females. Person correlation analysis was used to detect association between vitamin B_6 intake and plasma PLP concentration and urinary 4-PA execration. Differences and correlations were considered significant whenever the p-value was ($p \leq 0.05$).

RESULTS

Characteristics of participants are shown in Table 1. Means of height and weight were significantly lower in the female group compared to the male group. However, there was no significant difference between the two groups in the means of BMI. According to the classification of the National Health, Lung and Blood Institute (1998), the study found that in the male group, 63.8% (n = 81) of participants were overweight or obese and 34.6% (n = 44) were normal. In the female group, 66.1% (n = 74) of participants were overweight or obese and 30.4% (n = 34) were normal. Results of dietary intake are shown in Table 2. The female participants consumed less protein and vitamin B_6 compared to males, but the differences were not significant. The

Table 1: General characteristics among Adult Saudis in Al-Qassim Region-Saudi Arabia*

Characteristics	Males (n = 127)	Females (n = 112)
Age (yr)	28.7±8.8[a]	25.6±7.2[a]
Height (cm)	168.3±6.3[a]	156.2±5.9[b]
Weight (kg)	76.6±12.9[a]	63.2±8.8[b]
BMI (kg/m^2)	27.3±3.9[a]	26.7±4.6[a]

*Values are expressed as mean ± SD. [a, b]Different letters in a given row denote a significant differences, P-value ≤0.05.

Table 2: Intake of protein, vitamin B_6, vitamin B_6: protein ratio and percentage of vitamin B_6 from animal sources among Adult Saudis in Al-Qassim Region-Saudi Arabia*

Nutrient and source	Males (n = 127)	Females (n = 112)
Protein (g)	102.3±19.7[a]	96.6±20.6[a]
Vitamin B_6 (mg)	1.97±0.58[a]	1.89±0.61[a]
B_6: protein ration (mg g^{-1})	0.021±0.003[a]	0.020±0.04[a]
% of B_6 from animal sources	56.3±9.7[a]	38.9±10.4[b]

*Values are expressed as mean ± SD, [a, b]Different letters in a given row denote a significant differences, P-value ≤0.05.

percentage of vitamin B_6 from animal source was significantly lower in female group compared to male group. This indicates that males compared to females, consumed more meats and poultry and less cereals, legumes and vegetables. Major source of vitamin B_6 of both groups were mutton, chicken, banana and watermelon. Mean intakes of protein for both groups as well as for all participants from both groups were within the range of the AMDRs for protein (10-35% of energy), which was established in the DRI (2005). The mean intake of vitamin B_6 was adequate for both male and female groups, based on the recommended value of DRI (1998) (1.3 mg/day for adult male and female). None of the participants from either group had an average intake of vitamin B_6 below 1.3 mg/day. The biochemical indices of vitamin B_6 status are shown in Table 3. The means concentration of plasma PLP and means urinary 4-PA excretion of both group indicated an adequate status according to the suggested values for adequate status. These suggested values are $\geq$20 nmol L^{-1} (Lui *et al*., 1985) for plasma PLP and $\geq$0.3 μmol day^{-1} for urinary 4-PA excretion (Leklem, 2001). None of the participants of the two groups had plasma PLP concentration or urinary 4-PA excretion below the suggested values of adequacy. Comparison of males and females groups showed no significant difference in any of vitamin B_6 biochemical indices. Vitamin B_6 intake was significantly correlated with both plasma PLP concentration and urinary 4-PA excretion ($r = 0.37$, $p \geq 0.02$) ($r = 0.36$, $p \leq 0.02$), respectively. The mean albumin concentrations for both groups were within normal range (38-55 g L^{-1} for males and 35-52.5 g L^{-1} for females) (Gibson, 2005) with no significant difference between the two groups. Similarly, the means of plasma alkaline phosphatase activity for both groups were within the normal range (13-39 UL) (Lee and Nieman, 2003).

Table 3: Concentration of plasma PLP, Red Blood Cell (RBC) PLP and urinary 4-PA among Adult Saudis in Al-Qassim Region- Saudi Arabia*

Biochemical Indices	Males (n = 127)	Females (n = 112)
Plasma PLP (nmol L^{-1})	29.6±9.6[a]	31.7±10.4[a]
RBC PLP (nmol L^{-1})	26.9±5.8[a]	27.6±5[a]
Urinary 4-PA (μmol day^{-1})**	7.0±2.4[a]	6.8±2.5[a]

*Values are expressed as mean ± SD, **Average of three consecutive days, [a,b]Different letters in a given row denote a significant differences, P-value $\leq$0.05.

Table 4: Concentration of plasma albumin and alkaline phosphotase activity among Adult Saudis in Al-Qassim Region-Saudi Arabia*

Biochemical measurements	Males (n = 127)	Females (n = 112)
Plasma albumin (g L^{-1})	48.7±1.9[a]	49.1±2.0[a]
Plasma alkaline phosphatase (UL)	26.3±4.7[a]	24.2±3.8[a]

*Values are expressed as mean ± SD, [a,b]Different letters in a given row denote a significant differences, P-value $\leq$0.05.

DISCUSSION

The results of BMI showed prevalence of overweight and obesity among male and female participants. This is in agreement with several previous studies conducted in the Saudi Arabia population (Al-Nuaim *et al*., 1997; El-Hazmi and Warsy, 1997; Madani *et al*., 2000; Al-Assaf and Al-Numair, 2007). The aim of this study was not to evaluate obesity, but the importance of the BMI data is to indicate that the majority of participants were in the state of well or over nutrition. Thus, vitamin B_6 deficiency is not expected since signs of its deficiency due to dietary deficiency are rarely seen (Leklem, 2001; Dakshinamurti and Dakshinamurti, 2007). Dietary intake data showed high mean protein intake for both groups although they were within the AMDRs. This was due to the high intake of calories, which is in agreement with the data of the BMI mentioned earlier. Intake of protein and vitamin B_6 were adequate for both groups, which is in agreement with previous data (Al-Assaf and Al-Numair, 2007). The mean vitamin B_6 to protein ratio (Table 2) indicated an adequate vitamin B_6 status for both groups based on the suggested ratio reported by Leklem (1990) ($\geq$0.020). The mean % of vitamin B_6 from animal sources was significantly lower in females group compared to males group, which suggests lower bioavailability of the vitamin for females group compared to males group. Also, females group compared to males group had lower mean intake of vitamin B_6 and lower vitamin B_6: protein ratio (not significant). However, both plasma and RBC PLP concentrations were higher (not significant) in females group compared to males group. There are two explanations for this conflict. First, males compared to females consumed more protein, which may lower plasma PLP concentration. This relation was reported in previous studies (Miller *et al*., 1985; Hansen *et al*., 1996; Hansen *et al*., 1997). The second explanation is the prevalence of cigarette smoking among Saudi males as reported in several previous studies (Jarallah *et al*., 1999; Siddiquie *et al*., 2001; Al-Assaf, 2007). Several studies reported the adverse affect of cigarette smoking on plasma PLP concentration, which is in agreement with results of this study (Serfontein and Ubbink, 1988; Giraud *et al*., 1995; Giraud and Driskell, 1994). However, all of the indices of vitamin B_6 status indicated an adequate status for both groups. The mean plasma PLP concentration was significantly correlated with vitamin B_6 intake. This correlation is in agreement with previous reports (Chang *et al*., 2007; Kretsch *et al*., 1991; Huang *et al*., 1998). Also, the mean urinary 4-PA excretion was significantly correlated with vitamin B_6 intake, which is in agreement with previous data (Shultz and Leklem, 1981). There is no available suggested value of vitamin B_6 adequacy base on RBC PLP concentration. However, RBC PLP concentrations found in this study were lower than the concentrations reported by Reynolds *et al*.

(1988). The last mentioned study reported that a high intake of vitamin B_6 (2.11±0.12 mg/day) resulted in RBC PLP concentration of 85±6 nmol L^{-1}. Comparison the results of this study with other studies conducted in Saudi population is not possible since this is the first study investigated vitamin B_6 status among Saudi people. The normal mean of plasma alkaline phosphatase activity and plasma albumin concentration found in this study of both groups suggested no adverse affect on plasma PLP; thus no adverse affect on vitamin B_6 status as indicated by plasma PLP concentration. Results of the last two mentioned biochemical measurements are in agreement with normal ranges of adult Saudis reported by El-Hazmi *et al.* (1982) and Scott (1982).

Conclusion: In conclusion, results of this study showed that adult Saudi living in Al-Qassim region-Saudi Arabia had adequate status of vitamin B_6. This adequacy was evaluated by dietary measurements including vitamin B_6 intake, protein intake and vitamin B_6 to protein ratio. The adequacy was also measured by biochemical indices including plasma PLP concentration and urinary 4-PA excretion. This study suggests that, due to the numerous functions of vitamin B_6, further evaluations of the vitamin status are needed in various groups of Saudi population in other regions of the country.

REFERENCES

Al-Assaf, A.H., 2007. Anemia and iron intake of adult Saudis in Riyadh City-Saudi Arabia. Pak. J. Nutr., 6: 355-358.

Al-Assaf, A.H. and K.S. Al-Numair, 2007. Body mass index and dietary intake of Saudi adult males in the Riyadh region Saudi Arabia. Pak. J. Nutr., 6: 414-418.

Al-Jebrin, A.W.N. Sawaya, J.P. Salji, M. Ayaz and J.K. Khalil, 1985. Chemical and nutritional quality of some cereals and legumes based Saudi Arabian dishes II. Mineral and vitamin contents. Ecology Food And Nutr., 17: 345-352.

Al-Nuaim, A. E.A. Bamgboye, K.A. Al-Rubeaan and Y. Al-Mazrou, 1997. Overweight and obesity in Saudi Arabia adult population, role of socio-demographic variables. J. Community Health, 22: 211-223.

Al-Shoshan, A., 1992. The affluent diet and its consequences: Saudi Arabia- a case in point. World Rev. Nutr. Diet, 69: 113-165.

Bhagavan, H.N., 1985. Interaction between vitamin B_6 and drugs. In: Vitamin B_6: Its role in health and disease. Reynolds, R.D. and Leklem, J.E. (Eds). A.R. Liss, New York, NY., p: 401-415.

Chang, S.L. Hsiao, Y. Lee and S. Hsuen, 2007. Vitamin B_6 status assessment in relation to dietary intake in high school students aged 16-18. Br. J. Nutr., 97: 764-769.

Dakshinamurti, S. and K. Dakshinamurti, 2007. Vitamin B_6. In: Handbook of vitamins. Zempleni, J. *et al.* 4th Ed. CRC Press. Taylor and Francis Group. USA., p: 321-322.

DRI, 1998. Dietary Reference Intakes for thiamin, riboflavin, niacin, vitamin B_6, folate, vitamin B_{12}, pantothenic acid, biotin and choline. Washington DC: National Academy Press.

DRI, 2005. Dietary Reference Intakes for energy, carbohydrates, fiber, fat, fatty acids, cholesterol, protein and amino acids (macronutrients). Washington DC: national Academy Press.

El-Hazmi, M.A. and A.S. Warsy, 1997. Prevalence of obesity in the Saudi population. Ann. Saudi Med., 17: 302-306.

El-Hazmi, M.A., F.Z. Al-Faleh, I.A. Al-Mofleh, A.S. Warsy and A.K. Al-Askah, 1982. Establishment of normal reference ranges for biochemical parameters for healthy Saudi Arabs. Trop. Geogr. Med., 34: 323-332.

Food Processor plus computer program Software, 2001. (ESHA) Version 6; Salem, Oregon.

Gibson, R.S., 2005. Assessment of visceral protein status. In: principles of nutritional assessment. 2nd Ed. Oxford University Press, p: 410-412.

Giraud, D.W. and J.A. Driskell, 1994. Vitamin B_6 status of tobacco smokers, chewers and Non-users. Nutr. Res., 14: 1155-1164.

Giraud, D.W., H.D. Martin and J.A. Driskell, 1995. Erythrocyte and plasma B_6 vitamer concentrations of long-term tobacco smokers, chewers and nonusers. Am. J. Clin. Nutr., 62: 104-109.

Gregory, J.F. and J.R. Kirk, 1979. Determination of urinary 4-pyridoxic acid using high performance liquid chromatography. Am. J. Clin. Nutr., 32: 879-883.

Hansen, C.M., J.E. Leklem and L.T. Miller, 1996. Vitamin B_6 status indicators decrease in women consuming a diet high in pyridoxine glucoside. J. Nutr., 126: 2512-2518.

Hansen, C.M., J.E. Leklem and L.T. Miller, 1996. Vitamin B_6 status of women with a constant intake of vitamin B_6 changes with three levels of dietary protein. J. Nutr., 126: 1891-1901.

Hansen, C.M., J.E. Leklem and L.T. Miller, 1997. Changes in vitamin B_6 status indicators of women fed a constant protein diet with varying levels of vitamin B_6. Am. J. Clin. Nutr., 66: 1379-1387.

Huang, Y. W. Chen, M.A. Evans, M.E. Mitchell and T.D. Shultz, 1998. Vitamin B_6 requirement and status assessment of young women fed a high protein diet with various levels of vitamin B_6. Am. J. Clin. Nutr., 67: 208-220.

Jarallah, J.S., K.A. Al-Rubeaan, A. Al-Nuaim, A.A. Al-Ruhaily and K.A. Kalantan, 1999. Prevalence and determinants of smoking in three regions of Saudi Arabia. Tob. Control, 8: 53-56.

Kimura, M.K. Kanehira and K. Yokoi, 1996. Highly sensitive and simple liquid chromatographic determination in plasma of B_6 vitamins, especially pyridoxal-5-phosphate. J. Chromatagr A., 722: 269-301.

Kretsch, M.J., H.E. Sauberlich and E. Newburn, 1991. Electroencephalographic changes and periodontal status during short-term vitamin B_6 depletion of young, non-pregnant women. Am. J. Clin. Nutr., 53: 1266-1274.

Lee, R.D. and D.C. Nieman, 2003. Biochemical assessment of nutritional status. In: Nutritional assessment. 3rd Ed. McGrwa-Hill Companies. USA., p: 304-310.

Leklem, J.E., 1990. Vitamin B_6: status report. J. Nutr., 120: 73-77.

Leklem, J.E., 1999. Vitamin B_6. In: Modern Nutrition in Health and Disease. Shils, M.E.; Olson, J.A.; Shike, M. and Ross, A.C. (Eds) 9th Ed. Lippincott Williams and Wilkins. Baltimore, MD., p: 413-421.

Leklem, J.E., 2001. Vitamin B_6. In; Handbook of Vitamins. 3rd Ed. Machlin, L.J. Marcel-Dekker, Inc. NewYork, NY., p: 340-351.

Lui, A.L. Lumeng, Aronoff and T.K. Li, 1985. Relationship between body store of vitamin B_6 and plasma pyridoxal-P clearance: Metabolic balance studies in humans. J. Lab. Clin. Med., 106: 491-497.

Madani, K.A., N.S. Al-Amoudi and T.A. Kumosani, 2000. The state of nutrition in Saudi Arabia. Nutr. Health, 14: 17-31.

Miale, J.B., 1977. Cell washing. In: Laboratory medicine hematology: Serology and immunology, general laboratory technique. 5th Ed. By Amanda F, Cashen M.D. Tanya. M, Wildes M.D. The C.V. Mosby Company., p: 1047-1048.

Miladi, S.S., 1998. Changes in food consumption patterns in the Arab countries. Int. J. Food Sci. and Nutr., 49: S22-S30.

Miller, L.T., J.E. Leklem and T.D. Shultz, 1985. The effect of dietary protein on the metabolism of vitamin B_6 in humans. J. Nutr., 115: 1663-1672.

National Health, Lung and Blood Institute, 1998. Clinical guidelines on the identification, evaluation and treatment of overweight and obesity in adults. The evidence report. National Institute of Health. US Dept. of Health and Human Services. Obes Res., 1998 Sep;6 Suppl, 2: 51S-209S.

Reynolds, R.D., P.B. Moser-Veillon and A.K. Kant, 1988. Effect of age on status and metabolism of vitamin B_6 in men. In: Clinical and physiological applications of vitamin B_6. Leklem, J.E. and Reynolds, R.D. (Eds). Alan R. Liss. New York, NY., p: 109-125.

Reynolds, R.D., 1988. Bioavailability of vitamin B_6 from plant foods. Am. J. Clin. Nutr., 48: 863-867.

Roy, A.V., 1970. Rapid method for determining alkaline phospatase activity in serum with thymolpthalien mono-phosphate. Clin. Chem., 16: 431-436.

Sawaya, W.N., A. Al-Jebrin, S.P. Salji, M. Ayaz and J.K. Khalil, 1986. Nutritional evaluation of selected meat based Saudi dishes. Ecology of Food and Nutr., 18: 171-182.

Scott, T.G., 1982. Pilot study of the reference values for the commoner haematological and biochemical parameters in Saudi nationals. J. Clin. Pathol., 35: 69-73.

Serfontein, W.J. and J.B. Ubbink, 1988. Vitamin B_6 and myocardial infarction. In: Clinical and physiological applications of vitamin B_6. Leklem, J.E. and Reynolds, R.D. (Eds). Alan R. Liss, Inc. New York, NY, p: 201-217.

Shultz, T.D. and J.E. Leklem, 1981. Urinary 4-pyridoxic acid, urinary, vitamin B_6 and plasma pyrridoxal phosphate as measures of vitamin B_6 status and dietary intake of adults. In: Methods in vitamin B_6 nutrition. Leklem, J.E. and Reynolds, R.D. (Eds). Plenum Press. New York, NY., p: 297-320.

Siddiquie, S., D.O. Ogbeide and I. Al-Khaslifal, 2001. Smoking in a Saudi community: prevalence, influencing factors and risk perception. Fam. Med., 33: 367-370.

Slater, L., P.M. Carter and J.R. Hobbs, 1975. Measurement of albumin in the sera of patients. Ann. Clin. Biochem., 12: 33-38.

Vasilaki, A.T., D. McMillan, J. Kinsella, A. Duncan, D. O'Reilly and D. Talwar, 2008. Relation between pyridoxal and pyridoxal phosphate concentrations in plasma, red cells and white cells in patients with critical illness. Am. J. Clin. Nutr., 88: 140-146.