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ORIGINAL RESEARCH ARTICLE

Prevalence of Nutritional Anemia and Hyperhomocysteinemia in Urban Elderly

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Abstract The data on the prevalence of nutritional anemia among the urban elderly population in India was limited. Hence, the present study was carried out with an aim to assess the prevalence of nutritional anemia and its association with vitamin B12, folate, ferritin and homocystine among the urban elderly population. A community- based cross-sectional study was carried out among 282 urban elderly (≥ 60 years) subjects (186 males and 96 females) in Hyderabad. Fasting blood samples were collected and hemoglobin (Hb) was estimated by cyanmethemoglobin method. Plasma Folic acid and vitamin B12 levels were estimated by RIA and homocysteine and ferritin levels were estimated by ELISA methods. The overall prevalence of anemia (Hb < 12 g/dL for females and < 13 g/dL for males) among the urban elderly was 20.6% and the prevalence was found to be increasing with the age. The prevalence of vitamin B12 (< 203 pg/mL), folic acid (< 4 ng/mL), ferritin (< 15 ng/mL) and hyperhomocysteinemia (≥ 12 µmol/L) in these subjects was 36.0%, 8.2%, 1.1% and 24.3% respectively. The prevalence of anemia due to deficiencies of iron (ferritin < 15 ng/mL), folate and vitamin B12 was 5.45%, 9.1% and 42.3% respectively. A significant association was observed between the prevalence of anemia with ferritin and hyperhomocysteinemia. In conclusion, the prevalence of anemia and nutritional anemia among the urban-based elderly was 20.6% and 56.85% respectively. The association of anemia with hyperhomocysteinemia needs further studies.

Keywords Nutritional anemia · Elderly · Ferritin · Folic acid · Homocysteine · Vitamin B12

Introduction

The life expectancy of the world population has led to a progressive increase in an absolute number of elderly persons over the past 50 years. There has been an increase in elderly population (aged 60 years or above) in India from 77 million (7.7%) in 2001 to 104 million (8.6%) in 2011 [1]. This higher proportion of elderly population could be attributed to increased longevity, but is associated with various health problems including anemia. Anemia is a common health problem among the elderly and it affects the quality of individual life. Anemia is also associated with decreased functional ability/physical function [2], increased dementia [3], increased risk of falls, morbidity and mortality [4]. Since anemia negatively impacts on the overall well-being of elderly individuals, it is important to identify the factors associated with the anemia in order to overcome anemia related adverse health effects.

Anemia is multifactorial which includes both nutritional (nutrient-deficiencies) and non-nutritional (chronic diseases, chronic blood loss from the gastrointestinal tract, inflammation, unexplained anemia etc.) factors. About one-

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third of anemia in the elderly could be attributed to nutritional deficiencies of iron, folate, vitamin B12 or a combination of these three nutrients is termed as nutritional anemia [5]. Though the most common type of nutritional anemia is iron deficiency anemia (IDA), which accounts for nearly half of the nutrient deficiency-related anemia cases; the other two (vitamin B12, and folic acid) nutrient deficiencies also account for nearly 14% of all anemia cases in elderly [6]. Deficiency or low levels of vitamin B12 and folate increases homocysteine (Hcy) levels in the plasma [7] and is a known cardiovascular risk factor.

Though nearly one-third of the nutritional anemia in elderly is due to multiple nutrient deficiencies; earlier studies on prevalence of anemia in urban elderly was reported based on hemoglobin levels alone [8, 9]. Therefore, the present study was carried out with the aim to assess the prevalence of nutritional anemia among free-living urban elderly residing in Hyderabad, a metropolitan city in South India.

Materials and Methods

Subjects

This community-based cross-sections study was conducted in the year 2015 after obtaining clearance from Institutional Human Ethics Committee. Elderly people ($\geq$ 60 years) of both the genders were included in the study after obtaining written informed consent. Subjects below 60 years of age and those who are bedridden were excluded from the study. A total of 282 urban elderly people residing in Hyderabad were selected randomly. A pre-tested schedule was used for collecting information on the demographic profile and dietary habits.

The sample size was calculated based on the prevalence of anemia as 27% [10], considering 95% CI and 20% relative precision, a sample size of 260 was arrived. However, we covered a sample size of 282 elderly for this study.

Blood Sample Collection

Intravenous fasting blood samples were collected from elderly subjects in heparinized tubes and plasma samples were separated and stored at − 80 °C until further analysis. Before the separation of plasma, hemoglobin was estimated in whole blood samples by Cyanmethemoglobin method and the prevalence of anemia was calculated according to the WHO recommended cutoffs: Hemoglobin concentration below 12.0 g/dL in non-pregnant women and below 13.0 g/dL in men [11].

Biochemical Analysis

Vitamin B12 and folic acid in the plasma samples were determined by dual count Radioimmunoassay (RIA) kit (MP Biomedicals, NY, USA). The prevalence of vitamin B12 and folic acid deficiencies are assessed based on the WHO cutoffs [12]. Ferritin levels in plasma samples were estimated by sandwich ELISA as described earlier [13] and low iron stores was defined when serum concentrations of ferritin was < 15 µg/L [11]. Plasma homocysteine was estimated by ELISA (Cell Biolabs, San Diego, USA) and homocysteine levels ≥ 12 µmol/L was considered as hyperhomocysteinemia [14].

Vitamin B12 deficiency anemia is calculated as hemoglobin < 12 g/dL with plasma vitamin B12 < 203 pg/mL for women and with hemoglobin < 13 g/dL with plasma vitamin B12 < 203 pg/mL for men. Folic acid deficiency anemia is calculated as hemoglobin < 12 g/dL with plasma folic acid < 4.0 ng/mL for women and with hemoglobin < 13 g/dL with plasma folic acid < 4.0 ng/mL for men. Iron deficiency anemia is calculated as hemoglobin < 12 g/dL with plasma ferritin < 15 ng/mL for women and hemoglobin < 13 g/dL with plasma ferritin < 15 ng/mL for men [15].

Statistical Methods

Statistical analysis was performed by using the Statistical Package for Social Science (SPSS, 2005, version 19.0, Chicago). Data are presented as mean $\pm$ SE and/or prevalence (%). Mean values of haemoglobin, vitamin B12, folic acid, ferritin and homocysteine across the gender were compared by Students *t* test/Nonparametric of Mann–Whitney "*U*" test. To study the association between micronutrient deficiencies with anemia, the Chi square test was used and $p < 0.05$ was considered as significant.

Results

A total of 282 elderly subjects residing in Hyderabad metropolitan city were recruited. Of them, 186 (66.0%) were men and 96 (34.0%) were women and the mean age of the study subjects was 66.74 $\pm$ 0.34 years (60–93 years). In the age distribution, the majority of subjects (112) were in the age group of 60–64 years followed by 88 subjects in 65–69 years age group and 82 subjects were in the age group of 70 years and above. According to the dietary habits, about 61.0% of the study subjects were non-vegetarians and 39.0% subjects were vegetarians (Table 1).

The mean hemoglobin, plasma vitamin B12, folic acid, homocysteine, ferritin values are presented in Table 2.

Table 1 Particulars of urban elderly study subjects

Variable	n/Mean ± SE	Percentage
Total number of subjects	282	100
Male	186	65.95
Female	96	34.04
Age (years)	66.74 ± 0.34	N/A
60–64	112	39.7
65–69	88	31.2
≥ 70 years	82	29.1
Food habits (n = 260)		
Vegetarian	101	38.8
Non-vegetarian	159	61.2

n = number in parentheses; value is in mean ± SE; N/A = not applicable

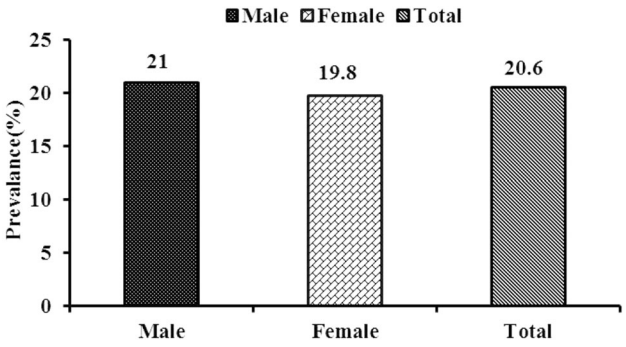

Fig. 1 Prevalence of anemia among urban elderly

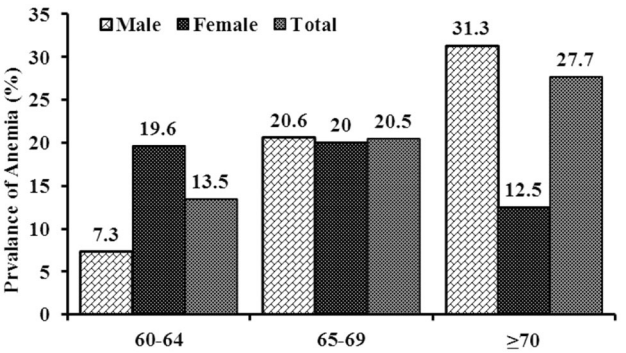

Fig. 2 Prevalence of anemia according to the age and gender in urban elderly

There was a significant ($p < 0.05$) difference in mean Hb and vitamin B12 levels between men and women. The mean folic acid levels were found to be high in elderly women as compared to elderly men. However, there was no significant difference in mean folic acid levels between genders. The levels of homocysteine was significantly ($p < 0.01$) higher among elderly women when compared to their male counterparts and significantly ($p < 0.05$) higher levels of ferritin was observed among elderly men when compared to women.

The overall prevalence of anemia in terms of haemoglobin among urban elderly was 20.6% and the corresponding figure among men and women was 21% and 19.8%, respectively (Fig. 1). The prevalence of anemia was found to be increasing with advancing age (Fig. 2). The prevalence of anemia in relation to food habits is shown in Table 3 and there was no significant difference in the prevalence of anemia between vegetarians (18.8%) and non-vegetarians (17.1%).

The overall prevalence of vitamin B12, folic acid, ferritin deficiencies and hyperhomocysteinemia was 36.0%, 8.2%, 1.1% and 24.3%, respectively (Fig. 3).The prevalence of vitamin B12 deficiency anemia was found in 22 (42.3%) subjects with anemia, while folic acid deficiency anemia was found in 5 (9.1%) subjects with anemia. Iron deficiency anemia was found to be in 3 (5.45%) subjects with anemia. In addition, an association of micronutrient deficiencies and dietary habits (vegetarian and non-vegetarian) with anemia is presented in Table 3. A significant association ($p < 0.05$) was found between elevated homocysteine levels, low ferritin levels with anemia. However, such association was not found between low vitamin-B12, Folic acid and dietary habits with anemia.

Table 2 Mean hemoglobin, vitamin B12, folic acid and homocysteine levels in urban elderly subjects

Particulars	Men	n	Women	n	Total	n
Hemoglobin (g/dL)*	14.54 ± 0.160	186	13.40 ± 0.23	96	14.15 ± 0.134	282
Vit-B12 (pg/mL)*	420 ± 39.4	180	464 ± 42.4	98	436 ± 29.5	278
Folic acid (ng/mL)	13.41 ± 0.93	182	14.7 ± 1.64	97	13.86 ± 0.83	279
HC (μmoles/L)*	9.14 ± 0.48	154	13.26 ± 1.52	85	10.61 ± 0.634	239
Ferritin (ng/mL)*	75.35 ± 3.14	182	61.63 ± 4.12	93	70.71 ± 2.53	275

Values are mean ± SE; *$p < 0.05$ (significant difference between gender); HC: Homocysteine; n = number in parentheses

Table 3 Association of anemia with vitamin B12, folic acid, homocysteine, ferritin and type of diet

Variables	N	Anemic (%)	Chi-square	*p*-value
Vitamin-B12 (pg/mL)				
< 203	98	22.4	1.099	0.294
≥ 203	174	17.2		
Folic acid (ng/mL)				
< 4	23	21.7	0.040	0.842
≥ 4	250	20.0		
Homocystine (μmol/L)				
< 12	177	13.6	6.086	0.014
≥ 12	58	27.6		
Ferritin (ng/mL)				
< 15	3	100.0	12.1	0.001
≥ 15	272	19.1		
Type of diet				
Vegetarian	101	16.8	0.001	0.975
Non-vegetarian	159	17.0		

N = number in parentheses

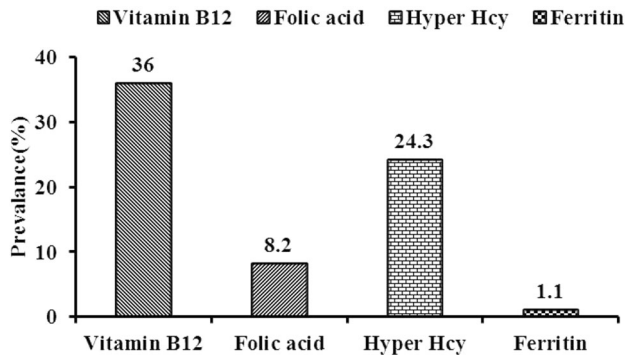

Fig. 3 Prevalence of vitamin B12, folic acid deficiencies, hyperhomocysteinemia (Hper Hcy) and ferritin deficiency in urban elderly

Discussion

In the present cross-sectional study, we studied the prevalence of nutritional anemia in south Indian free-living urban elderly subjects. The results of the current study revealed that the overall prevalence of anemia (based on Hb) was 20.6% which is comparable with study carried out in Mysore [16]. Nevertheless, the present study reported the prevalence of anemia is low when compared to the prevalence (45.5%) reported among community-dwelling elderly of Assam [8]. The lower prevalence of anemia in our study may be due to the consumption of foods rich in micronutrients or nutrition supplements. In addition, the prevalence of anemia in the present free-living urban elderly subjects was low when compared to the hospital based study in India where, they reported the prevalence was 37.9% [17].

The current prevalence of anemia of 20.6% is higher than the studies reported in community dwelling elderly in Ankara (Turkey) and Beijing (China) with the prevalence of 7.3% and 14.8% respectively [15, 18]. Data from the United States population assessed during the Third National Health and Nutrition Examination Survey (NHANES III) reported the prevalence of anemia in people aged > 65 years was 10.6% [5] which was low when compared to our current study. Nevertheless, the present study finding, the prevalence of anemia is comparable with the prevalence of 18.6% from southeastern Brazilians [19]. These variations in the prevalence of anemia in different countries could be due to different geographical and dietary variations, and also usage of difference age cutoffs (> 60 years and > 65 years) in different studies.

Anemia is a multi-factorial condition in elderly and its prevalence usually increases with age particularly after 60 years and the same was reported by the earlier population-based studies [5, 8, 20]. The findings of the present study are also consistent with these studies as there was a notable difference in the prevalence of anemia in the age groups 60–64 years, 65–70 years and > 70 years.

According to the previous study [21], type of diet may also influence the prevalence of anemia. Although, in our study we did not find much difference in the prevalence of anemia between vegetarians (18.8%) and non-vegetarians (17.1%), a recent study conducted among community-dwelling elderly of Assam reported that the prevalence of anemia was high (53%) among vegetarians as compared to the non-vegetarians (39%) [8] and this could be due to the difference in quantity and frequencies of consumption of animal source foods.

Elderly people are more vulnerable for micronutrient deficiencies especially with respect to iron, folate and vitamin B12. In the present study, the overall prevalence of vitamin B12 deficiency (36%) was high when compared to the prevalence of folate deficiency (8.2%) and hyperhomocysteinemia (24.3%). The current reported prevalence of folate deficiency was low and vitamin B12 deficiency was high when compared to earlier study reported by Sivaprasad et al. [10]. This present prevalence of vitamin B12, folate deficiencies and hyperhomocysteinemia was high when compared to the previous study [22]. These findings along with previous studies [7, 23] indicate that low levels of vitamin B12 could contribute to elevated levels of homocysteine and our study findings are in well agreement with these study reports, where we found high vitamin B12 deficiency with higher homocystine levels.

In the current study, the prevalence of anemia due to iron deficiency (based on ferritin levels < 15 ng/mL) was 5.45% which is marginally high when compared to the

prevalence (3.8% in 2006 and 2.2% in 2012) reported in Mexico and 2.8% in Turkey [15, 24]. The current prevalence was very low when compared to the prevalence of 48.3% reported in US older adults (> 65 years) [5].This marked difference in prevalence of IDA may be due to adoption of different diagnostic criteria by the different investigators and also due to wide range of ferritin cutoffs used in different studies.

The prevalence of anemia due to folate deficiency in the current study was 9.1%, which is higher when compared to the prevalence (1%) reported in elderly of Turkey [15] and US (6.4%) [5]. Interestingly, the prevalence of anemia due to vitamin B12 deficiency was 41.2% which is high when compared to the other nutrients (iron and folate) deficiency anemia and also high when compared to the other studies reported in US older adults > 65 years with the prevalence of 17.2% [5], and in Turkey with the prevalence of 4.4% [15]. The overall nutrient deficiency anemia in the present study was around 56% which is high as compared to the hospital based study in Indian elderly (47.6%) [25] and free-living US elderly (34.1%) [5]. The remaining unexplained anemia of 44% could be contributed to the other factors like erythropoietin insufficiency, cytokine inhibition of erythropoiesis, androgen decline, stem cell function and myelodysplasia [5, 26].

Further, we investigated the association of vitamin B12, folic acid, ferritin deficiencies and hyperhomocysteinemia with anemia. A very interesting finding of our study is that there is a significant association of anemia with ferritin deficiency ($p < 0.001$) and elevated homocysteine levels ($p < 0.014$), but not with vitamin B12 and folic acid deficiencies. This current observation was in line with the results of the previous study where it was reported that association of low hemoglobin levels with folate deficiency and elevated homocysteine levels in the elderly of > 85 years [27]. In another study conducted in Australia in older adults (> 49 years) as a part of Blue Mountains Eye Study (BMES), were observed similar findings that vitamin B12 deficiency and hyperhomocysteinemia was associated with prevalence of anemia [28]. In overall, the common observation in all the above three studies was association of anemia with hyperhomocysteinemia.

Conclusions

According to WHO definition of anemia, the current study revealed that the prevalence of anemia is 20.6% among the urban elderly participants which increases with age. The prevalence of anemia is associated with ferritin deficiency and elevated homocysteine levels but not with vitamin B12 and folic acid deficiencies. Anemia among the elderly is not simply a medical disorder but it adversely affects the quality of one's life. Thus, it may become a financial liability to the government as more resources may have to be allocated to correct the problem. Therefore, the underlying cause of anemia should be identified early in elderly and treated effectively.

Limitations

The inclusion of elderly subjects taking iron supplements and B-complex vitamins may have underestimated the prevalence of anemia in the present study.

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Compliance with Ethical Standards

Conflict of interest The authors declare that they have no competing interests.

References

1. Elderly in India-Profile and Programmes, 2016. http://www.mospi.gov.in/sites/default/files/publication_reports/ElderlyinIndia_2016.pdf.
2. Denny SD, Kuchibhatla MN, Cohen HJ. Impact of anemia on mortality, cognition, and function in community-dwelling elderly. Am J Med. 2006;119(4):327–34.
3. Hong CH, Falvey C, Harris TB, Simonsick EM, Satterfield S, Ferrucci L, et al. Anemia and risk of dementia in older adults: findings from the Health ABC study. Neurology. 2013;81(6):528–33.
4. Eisenstaedt R, Penninx BW, Woodman RC. Anemia in the elderly: current understanding and emerging concepts. Blood Rev. 2006;20(4):213–26.
5. Guralnik JM, Eisenstaedt RS, Ferrucci L, Klein HG, Woodman RC. Prevalence of anemia in persons 65 years and older in the United States: evidence for a high rate of unexplained anemia. Blood. 2004;104(8):2263–8.
6. Patel KV. Epidemiology of anemia in older adults. SeminHematol. 2008;45(4):210–7.
7. Yajnik CS, Deshpande SS, Lubree HG, Naik SS, Bhat DS, Uradey BS, et al. Vitamin B12 deficiency and hyperhomocysteinemia in rural and urban Indians. J Assoc Physicians India. 2006;54:775–82.
8. Agarwalla R, Saikia AM, Parashar M, Pathak R, Islam F. Assessment of prevalence of anemia in and its correlates among community dwelling elderly of Assam, India: a cross sectional study. Int J Nutr Pharmacol Neurol Dis. 2016;6(1):23–7.
9. Swami HM, Bhatia V, Dutt R, Bhatia SPS. A community based study of the morbidity profile among the elderly in Chandigarh, India. Bahrain Medi Bull. 2002;24(1):13–6.
10. Sivaprasad M, Shalini T, Balakrishna N, Sudarshan M, Lopamudra P, Suryanarayana P, et al. Status of vitamin B12 and folate among the urban adult population in South India. Ann Nutr Metab. 2016;68(2):94–102.

11. World Health Organization. Iron deficiency anaemia: assessment, prevention and control. A guide for programme managers. Geneva: WHO; 2001.
12. deBenoist B. Conclusions of a WHO technical consultation on folate and vitamin B12 deficiencies. Food Nutr Bull. 2008;29:S238–44.
13. Augustine LF, Nair KM, Rao SF, Rao MV, Ravinder P, Laxmaiah A. Exploring the bio-behavioural link between stress, allostatic load and micronutrient status: a cross-sectional study among adolescent boys. Indian J Med Res. 2016;144(3):378–84.
14. Satyanarayana A, Balakrishna N, Pitla S, Reddy PY, Mudili S, Lopamudra P, et al. Status of B-vitamins and homocysteine in diabetic retinopathy: association with vitamin-B12 deficiency and hyperhomocysteinemia. PLoS ONE. 2011;6(11):e26747.
15. Yildirim T, Yalcin A, Atmis V, Cengiz OK, Aras S, Varlı M, et al. The prevalence of anemia, iron, vitamin B12, and folic acid deficiencies in community dwelling elderly in Ankara, Turkey. Arch Gerontol Geriatr. 2015;60(2):344–8.
16. Shraddha K, Prashantha B, Prakash B. Study on morbidity pattern among elderly in urban population of Mysore, Karnataka, India. Int J Med Biomed Res. 2012;1(3):215–23.
17. Alwar V, Reethi K, Rameshkumar K. Geriatric anemia: an Indian perspective. Indian J Hematol Blood Transfus. 2013;29(2):126–7.
18. Ning SY, Chang NB, Han XY, Liu X, DuanYW Liu YH, et al. The prevalence and etiology of anemia in urban community dwelling elderly population in Beijing. Chin J Intern Med. 2016;55(04):289–92.
19. Callera F, Callera AF, da Silva AM, Rosa ES. Prevalence of anemia in a sample of elderly southeastern Brazilians. Rev Bras HematolHemoter. 2015;37(1):43–7.
20. Choi CW, Lee J, Park KH, Yoon SY, Choi IK, Oh SC, et al. Prevalence and characteristics of anemia in the elderly: cross-sectional study of three urban Korean population samples. Am J Hematol. 2004;77(1):26–30.
21. Mahajani Kamla, Bhatnagar Vibha. Comparative study of prevalence of anaemia in vegetarian and non vegetarian women of Udaipur City, Rajasthan. J Nutr Food Sci. 2015;S3:1–6.
22. Shobha V, Tarey SD, Singh RG, Shetty P, Unni US, Srinivasan K, et al. Vitamin B12 deficiency and levels of metabolites in an apparently normal urban south Indian elderly population. Indian J Med Res. 2011;134(4):432–9.
23. Mahalle N, Kulkarni MV, Garg MK, Naik SS. Vitamin B12 deficiency and hyperhomocysteinemia as correlates of cardiovascular risk factors in Indian subjects with coronary artery disease. J Cardiol. 2013;61(4):289–94.
24. Contreras-Manzano A, Cruz Vde L, Villalpando S, Rebollar R, Shamah-Levy T. Anemia and iron deficiency in Mexican elderly population. Results from the Ensanut 2012. Salud Publica Mex. 2015;57(5):394–402.
25. Raina A, Kumarb A, Singh A, Gupta G, Malhotra P, Raina SK. A clinicohaematological profile of elderly patientsbeing investigated for anaemia in a tertiary care centre in north-west India. Egypt J Haematol. 2014;39:190–4.
26. Makipour S, Kanapuru B, Ershler WB. Unexplained anemia in the elderly. Semin Hematol. 2008;45(4):250–4.
27. denElzen WP, Westendorp RG, Frolich M, de Ruijter W, Assendelft WJ, Gussekloo J. Vitamin B12 and folate and the risk of anemia in old age: the Leiden 85-Plus Study. Arch Intern Med. 2008;168(20):2238–44.
28. Gopinath B, Rochtchina E, Flood V, Mitchell P. Association of elevated homocysteine level and vitamin B12 deficiency with anemia in older adults. Arch Intern Med. 2009;169(9):901–2.