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Folate deficiency among aged patients and its amelioration through consumption of folic acid-fortified rice

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This is an observational study on the patients in a geriatric health care facility who received folic acid-fortified rice. Folate deficiency was prevalent among the patients in the rate of 38% at the beginning. The folate deficient states were ameliorated by the folic acid-supplementation within six months. The elevated levels of serum folate improved the red blood cell parameters. Another benefit of folic acid-fortified rice was that fever events were reduced in the patients with higher folate levels after folic acid-supplementation.

Key Words : folic acid, elderly patient, anemia, infection

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Introduction

Anemia is an issue of concern in the health management of older people. Anemia often signifies underlying diseases and is associated with poor clinical outcomes. According to the criteria by the World Health Organization, anemia is defined as hemoglobin concentration below 12.0 g/dL in women and below 13.0 g/dL in men (World Health Organization, 1968). Anemia is quite frequent in the aged people, as examined in Japan for community dwelling (Ohhara et al., 1990; Uno, 2010), institutionalized (Uno, 2010) and hospitalized elderly (Nitta et al., 1979; Hamaguchi et al., 2009; Hagino et al., 2009). Causes for anemia are clinically classified into groups such as bleeding, iron-deficiency, vitamin B12 deficiency, folate deficiency, chronic diseases, and unexplained causes.

In our geriatric facility for frail aged, a long-term care health facility, in order to explore the causes for anemia the serum concentrations of iron, vitamin B12 and folate were determined. The examination revealed that unexpectedly high prevalence of folate deficiency existed among elderly patients. The folate deficiency is well known to be another cause for macrocytic anemia or pernicious anemia rather than vitamin B12 deficiency (Wyngaarden & Smith, 1988), and suggested to be related with decreased immunity (Hamer et al., 2009). Furthermore, the folate deficiency is known to induce hyperhomocysteinemia that is supposed to be a risk for atherosclerosis (Giles, Kittner, Anda, Croft, & Casper, 1995; SEARCH Collaborative Group, 2010; Wald, Morris, & Wald, 2011), dementia (Kageyama, Hiraoka, & Kagawa, 2008; Seshadri et al., 2002), cancer (Phelip, Ducros, Faucheron, Flourie, & Roblin, 2008; Collin et al., 2010), and osteoporosis (Hagino et al., 2009; McLean, Jacques, Selhub, Fredman, Tucker, Samelson, Kiel, Cupples, & Hannan, 2008). To prevent such disease states due to decreased folate, strategies to increase the serum folate levels have been tried in the literature. In this study, folic acid-fortified rice was employed for the purpose. The grain product has been provided in the Japanese market by House Wellness Foods Co. (Itami, Hyogo) since March, 2010. This rice product was added to ordinary rice so that 220 μg of folic acid as monoglutamyl folic acid was given to each patient per day. This nutrient supplementation elevated the serum folate levels in most of patients, and ameliorated all the folate deficient states. This paper reports two beneficial effects observed on geriatric patients due to the folic acid-supplementation, that is, normalized blood cell sizes and reduced infectious events.

Methods

Subjects

Subjects enrolled in this study were inpatients in a long-term care health facility, Soka Royal Care Center (Soka city, Saitama prefecture of Japan). Most of them were older than 65 years of age and several percentages of them were 64 years of age or younger. Patients aged 65 years or more suffered from impaired activity of daily living to various degrees with or without chronic diseases. The degree of disability of individuals in need of care was assessed to be between 1 and 5 of nursing level according to the long-term care health insurance law in Japan. The averaged score of nursing level was 3.6 for all the patients. Patients younger than 64 years of age mostly suffered from neurological damages due to cerebral hemorrhage. Patients older than 65 years of age (100 women and 44 men) were examined for blood cell parameters. Parts of those were subjected to micronutrient assays and/or follow-up studies on the effects of folic acid-supplementation (Table 1). Patients who had kidney diseases or liver diseases were not included in this study.

Table 1. Subjects enrolled for blood tests and observation on the effects of folic acid-supplementation.

For blood tests	Subjects		Results shown in
	Number	Periods of study	
Blood cell parameters	144	2011	Table 2 and Figure 1
Micronutrient assays	68	2010	Table 3

For follow-up observations on the effects of folic acid-supplementation	Subjects		Results shown in
	Number	Periods of study	
Changes in serum folate levels	53	2010.1～2011.1	Figure 2
Changes in MCV* of red blood cells	21	2010.1～2011.1	Figure 3
Changes in fever event incidence	49	2009.4～2011.9	Figure 4

* Mean corpuscular volume

Supplementation of folic acid

Institutionalized patients were supplied meals in which the reference daily intake of nutrients were calculated by a national registered dietitian. Three meals a day were prepared to contain 240 μg of folate derived from food stuff on average. Additional supplementation of folic acid was made by adding calculated amounts of synthetic monoglutamyl folic acid-fortified rice (House Wellness Foods Co.,) to washed rice before cooking. We planned to add 220 μg folic acid to rice per person a day, expecting the serum folate levels to be elevated by 5.65 ng/mL as calculated using the equation of Quinlivan and Gregory (Dary, 2009).

Laboratory analysis

Blood samples were sent to Itabashi Medical Laboratory (Eil Co., Itabashi-ku, Tokyo) for analysis. Blood tests were performed before the folic acid-supplementation and thereafter at one- to three-month intervals during the supplementation period. Blood cell counts and the concentrations of hemoglobin, iron, vitamin B12 and folate were measured by instrumental analyses. Reference values for hemoglobin were >12.0 g/dL for women and >13.0 g/dL for men; those for iron were 50-160 μg/dL for women and 60-200 μg/dL; those for vitamin B12 were 233-914 pg/mL for both sexes; and those for folate were >4.0 ng/mL for both sexes.

Results

A total of 144 patients were subjected to blood tests, and the results are shown in Figure 1. A total of 144 patients were subjected to blood tests, and the results are shown in Figure 1. Red blood cell counts and hemoglobin concentrations were decreasing with advancing age (Figures 1A and 1B). The changing patterns in hemoglobin concentrations with age were very similar to those of serum albumin concentrations (Figure 1C). The prevalence of anemia was becoming higher in older age groups (Table 2). The anemic prevalence in men appeared much higher than that in women as comparing both the prevalence in the same age groups. The presence of high percentages of patients with high MCV (mean corpuscular volume) means high prevalence of macrocytic anemia in elderly

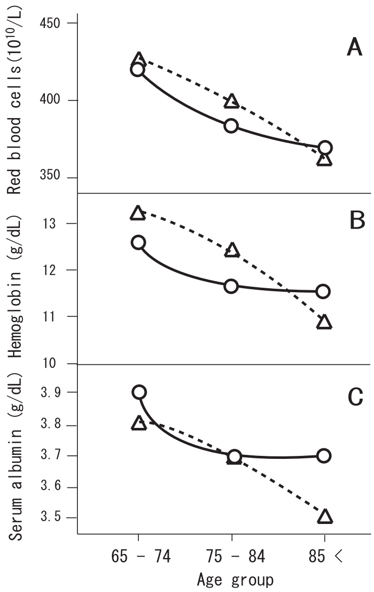

Figure 1. Red blood cells (A) and the concentrations of hemoglobin (B) and serum albumin (C) in three age groups of patients

old (65 – 74 years of age), older old (75 – 84), and oldest old (over 85). Open circles indicate the values for women and open triangles those for men.

patients. Patients with low albuminemia comprised more than 50% of the total. About one third of the patients were revealed to have both anemia and low albuminemia.

Table 2. Prevalence of anemia and low albuminemia status in patients

Blood status \ Age group	65 – 74 years Women (n=13) Men (n=9)	75 – 84 years Women (n=38) Men (n=26)	Over 85 years Women (n=49) Men (n=9)	Total Women (n=100) Men (n=44)
Anemia*1				
Women	7.7 %	52.6 %	57.1 %	49.0 %
Men	62.5 %	59.3 %	88.9 %	65.9 %
Both sexes	28.6 %	55.4 %	62.0 %	54.2 %
High MCV*2				
Women	30.8 %	50.0 %	63.2 %	54.0 %
Men	44.4 %	57.7 %	33.3 %	50.0 %
Both sexes	36.4 %	53.1 %	56.9 %	52.1 %
Low albuminemia*3				
Women	23.0 %	52.6 %	61.2 %	53.0 %
Men	44.4 %	50.0 %	66.7 %	52.3 %
Both sexes	31.8 %	51.6 %	62.1 %	52.8 %
Anemia with low albuminemia				
Women	7.7 %	36.8 %	36.7 %	33.0 %
Men	22.2 %	38.5 %	55.6 %	38.6 %
Both sexes	13.6 %	37.5 %	41.4 %	35.4 %

*1, Anemia was defined according to WHO criteria. The hemoglobin concentrations below 12.0 g/dL in women, and below 13.0 g/dL in men were assigned as anemia.
*2, MCV above 93 fL was defined as macrocytic status.
*3, Serum albumin below 3.8 g/dL was defined as low albuminemia.

About a half of the total patients were subjected for the analysis of the concentrations of iron, vitaminB12 and folate in serum (Table 3). The iron deficiency and vitamin B12 deficiency were present in the groups older than 75 years. On the other hand, folate deficiency was seen widely through presenile to oldest groups. The prevalence of folate deficiency was much greater than that of iron deficiency or vitamin B12 deficiency.

Patients (n=53) who stayed for longer periods than 6 months were enrolled for the folic acid-supplementation trial. Figure 2A shows the basal data on serum folate level distribution before the folic acid-supplementation, indicating large percentages of patients as about 40% to be assigned as folate deficiency. After one month of the supplementation, the serum folate distribution pattern largely shifted rightwards (Figure 2B). While the folic acid-supplementation was continued further, the serum folate levels were gradually increasing (Figures 2C and 2D). At the time of 6 months, all the patients became free of folate deficiency.

Table 3. Prevalence of patients with micronutrient deficiency in relation to possible causes for anemia

Age group / Patients	< 64 (n = 4)	65 – 74 (n = 13)	75 – 84 (n = 26)	85 < (n = 25)	Total (%) (n = 68)
Iron-deficient	0	0	3	5	8 (12%)
Vitamin B12-deficient	0	0	3	3	6 (9%)
Folate-deficient	3	7	7	9	26 (38%)
Sum	3	7	13	17	40 (59%)

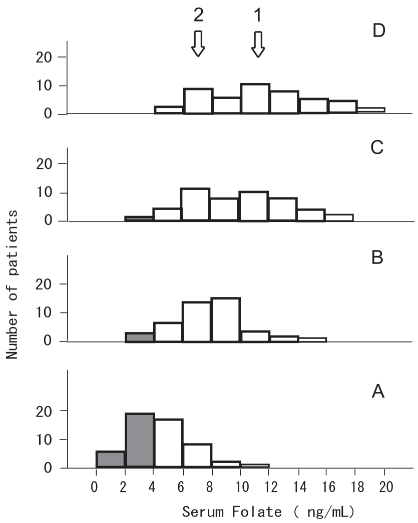

Figure 2. Serum folate distribution patterns shifted rightwards during the folic acid-supplementation for 6 months.

Patients (n=53) have taken 220 μg folic acid-containing rice per day. A, basal levels of serum folate; B, serum folate distribution patterns after one month-supplementation; C, serum folate distribution patterns after three month-supplementation; and D, serum folate distribution patterns after 6 month-supplementation. ●● columns indicate lower serum folate levels than 4.0 μg/mL that is the lower limit of reference level. Arrows show apparent two peaks (peak 1, good responder and peak 2, poor responder). Average values of serum folate are 4.1±2.0 μg and the median values are 4.4 μg in A, 7.9±3.0 μg and 7.6 μg in B, 10.1±3.4 μg and 10.0 μg in C, and 11.3±3.8 μg and 11.5 μg in D.

Looking at the pattern changes of serum folate level distribution, we noticed that the distribution patterns were composed of at least two components (Figures 2C and 2D). That is, one peak in Figure 2B seemed to tend to separate into two peaks with time as seen in Figures 2C and 2D. This may imply that there were at least two patient groups, a group of good responders and that of poor responders. Even poor responders recovered completely from folate deficient states in the course of receiving supplemental amounts of folic acid as 220 μg per day.

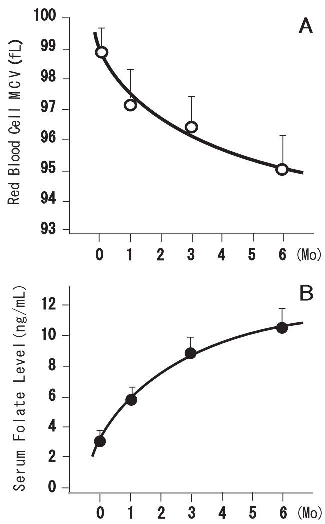

Figure 3. Changes in red blood cell MCV (A) and serum folate levels (B) during folic acid-supplementation

Folic acid-deficient patients (n=21). Values are expressed as average ± SE. MCV, mean corpuscular volume.

Patients with folate deficiency were found to be 26 (38%) out of 68 in Table 3. Five patients who had pernicious anemia among the folate deficient were subjected to medical treatment with 5000 μg of folic acid per day. Their folate levels jumped up from 2.8 ng/mL at the beginning to more than 400 ng/mL at one month of the treatment. Blood tests showed that MCV (mean corpuscular volume) values were lowered from 104.8 to 97.8 in average, and hemoglobin concentrations were increased from 10.6 to 12.5 g/dL in average during the 6 month treatment. Thus, the macrocytic anemia was greatly improved by the folic acid-treatment. The other 21 patients under folate deficient states were allocated to the group of folic acid-supplementation and monitored for a red cell parameter, MCV, during the supplementation. They showed higher MCV values than the upper reference range (93 fL) at the beginning, and the MCV values were decreasing inversely with the increasing serum folate levels for 6 months (Figure 3).

Among the elderly, especially patients in geriatric facilities, fevers are common symptoms due to various infectious attacks. The incidence of fever events was recorded for each patient, and the frequency since the folic acid-supplementation was begun was compared with that prior to the supplementation. Patients (n=49) who stayed for more than two years were enrolled, and were divided into two groups according to their basal values of serum folate, low folate (1.3-4.0 ng/mL; average, 2.7±0.8 ng/mL; n=24) and high folate groups (4.1-10.5 ng/mL; average, 5.9±1.7 ng/mL; n=25). Figure 4 shows the changes in the incidence of fever events in the two groups before and after the folic acid-supplementation. While both the groups showed similar incidence patterns before the supplementation, the high folate group appeared to greatly escape from fever events after the supplementation.

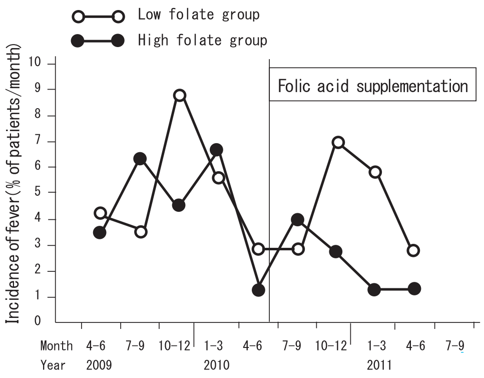

Figure 4. Changes in the incidence of fever events in the two groups, low folate and high folate groups of aged patients after folic acid-supplementation.

The average values of serum folate were 2.7±0.8 μg for the low folate group and 5.9±1.7 μg for high folate group in the begininng. The incidence is quarterly shown.

Discussion

Anemia is common in older people, and the prevalence of anemia is known to increase with advancing age (Beghe, Wilson, & Ershler, 2004; Takasaki, Tsurumi, Konjiki, Sakurai, Kanou, Yanagawa, & Katsunuma, 1997). The prevalence of anemia in elderly cohorts has been investigated among many countries including Japan. A systematic review on the anemia prevalence in older persons retrieved 45 studies, and summarized the results using a meta-analysis, that is, 12 % (8-25 %) anemia in studies based in the community, 47 % (31-50 %) in nursing homes and 40 % (40-72 %) in hospital admissions (Gaskell, Derry, Moore, & McQuay, 2008). In Japan, the prevalence of anemia in the elderly was reported to be 6.4 % for a healthy group and 52 % for an institutionalized group (Uno, 2010). Anemia among hospitalized aged patients was observed in a little higher rates such as 57 % (Hamaguchi et al., 2009) and 68 % (Hagino et al., 2009). In our geriatric facility, 54 % of patients had anemia (Table 2). This prevalence is comparable with a recently reported data (Uno, 2010).

The results of blood tests on a total of 144 patients are presented in Figure 1 and Table 2. The data are shown for stratified groups, old (65-74 years), older old (75-84) and oldest old (over 85). Three parameters, red cell counts, hemoglobin concentrations and albumin levels all were decreased with advancing age (Figure 1). The latter two parameters show very similar age changes, and may reflect decreasing hematopoiesis and nutritional states along with aging. Anemia states were found more in men than women in the same age groups (Table 2). This may relate with the shorter average life span of men than that of women.

As shown in Table 2, higher values of MCV (mean corpuscular volume) than 93 fL indicating red blood cells to be macrocytic are seen in 52 % of the patients. MCV of elderly persons is known to increase with advancing age (Nitta et al., 1979; Takasaki et al., 1997). The reason for that is implicated in deteriorating hematopoietic function (Takasaki et al., 1997). The reduction of hematopoiesis is thought to be partly caused by insufficiency or deficiency of B-group vitamins such as vitamin B12 and folic acid, because they are required for the erythroblast cell proliferation in the marrow (Wyngaarden, & Smith, 1988). Based on this point, we examined the concentrations of vitamin B12 and folate in serum in addition to iron, which is known to be a major cause for anemia (Table 3). The prevalence of folate deficiency was found to be unexpectedly high in 38 % of the patients examined in our geriatric facility. The prevalence of folate deficiency was reported to be 5 % among people aged 65-74 years and 10 % or higher among people aged 75 years or greater in the community of the United Kingdom (Clarke et al., 2004). As to the hospitalized elderly, the folate deficiency was reported to be 17 % by an old study (Nitta et al. 1979). The present data shows that the prevalence of folate deficiency is much higher than that of iron or vitamin B12 deficiency. This might indicate an aspect of malnutritional status of the diseased elderly who were institutionalized.

In order to correct the folate deficient states, we employed a method for nutritional supplementation to meals. Folic acid-fortified rice was used to enrich boiled rice or rice gruel with calculated amounts of folic acid so that 220 μg of folic acid could be provided to each patient per day. This amount of folic acid supplemented is about comparable with those reported in various folic acid supplementation trials (Hiraoka et al., 2009; Jacques , Selhub, Bostom, Wilson, & Rosenberg, 1999; Kauwell et al., 2000; Pitkin, 2007; Quinlivan & Gregory, 2003; Riddell, Chisholm, Williams & Mann, 2000; Venn et al., 2002; Ward et al., 1997).

Before and during the supplementation period, blood tests were performed. As shown in Figure 2, the number of folate deficient patients was largely reduced in a month, and all the deficient cases disappeared at the sixth month of the supplementation. According to our initial expectation, the serum folate levels should have sharply increased and reached some steady state levels in a short time, because folic acid is water-soluble and it is thought to be rapidly distributed and equilibrated in the body. However, this was not the case. Serum folate levels were rather slowly increasing. It was reported that a plateau in serum folate levels was achieved within 6 weeks when 200 μg of folic acid per day was dosed (Ward et al., 1997). This may not be true. In our trial, even after three months of folic acid-supplementation the serum levels of folate continued to increase as shown in Figure 2. Another point to be noticed was that the increasing rates were not consistent among patients. After the third month of the supplementation (Figures 2C and D), the distribution pattern appeared to be separated into at least two peaks, a right peak indicating a group of good responders at about 11 ng/mL and a left peak indicating a group of poor responders at about 7 ng/mL in Figure 2D. This might result from the different genetic backgrounds of patients. The C677T variant methylenetetrahydrofolate reductase (MTHFR), a key enzyme in the methylation of homocysteine to methionine, is supposed to be a cause of hyperhomocysteinemia which is known to be closely related with low serum folate status (Russo et al., 2003). The frequency of TT homozygote of MTHFR was estimated to be 17.3 % in Japanese women who showed poor responses to folic acid supplementation (Hiraoka, Kato, Saito, Yasuda, & Kagawa, 2004). Such the genetic polymorphism may underlie the different responses to folic acid-supplementation in the present study, where poor responders might be related to TT homozygotes and good ones to CC or CT. Our long-term supplementation trial reveals that the folate status of even poor responders can gradually be ameliorated by taking a low amount of folic acid as 220 μg per day for several months.

Patients with folate deficiency were found to show higher values of MCV (98.8 fL in average; n=21), indicating a macrocytic status. The MCV values were decreasing in accordance with increasing levels of serum folate due to the supplementation of 220 μg folic acid per day (Figure 3). The other patients with severe folate deficiency who had much higher values of MCV (104.8 fL in average; n=5) were subjected to medication by giving a tablet of 5000 μg folic acid per day. Their folate levels were elevated to more than 400 ng/mL and MCV values were down to 97.8 fL in average. Their hemoglobin concentrations were increased from 10.6 to 12.5 g/dL in average. Thus, the macrocytic status was ameliorated by the supplementation of small amounts of folic acid for modest folate deficient patients or administration of folic acid tablets to severe folate deficient ones.

Folate deficiency has been implicated in various adverse outcomes (Reynolds, 2002) in addition to macrocytic anemia or pernicious anemia. In 1990s, women of childbearing age with low serum folate were thought to have a risk of inborn errors such as neural-tube defects in their offsprings, and mandatory food fortification with folic acid was initiated in 1998 in the United States and Canada (Quinlivan & Gregory, 2003; De Wals et al., 2007) followed by United Kingdom (Reynolds, 2002) and other countries. After the food fortification with folic acid was implemented, the prevalence of neural-tube defects was reported to decrease by 46 % in Canada (De Wals et al., 2007). For elderly people, low folate status was correlated with senile dementia (Kageyama et al., 2008). An observational study showed that higher serum folate levels were associated with better cognitive function (De Lau, Refsum, Smith, Johston, & Breteler, 2007). The connection of folate deficiency to depression is suggested based on a probable mechanism linking methylation in the metabolism of catecholamines in the brain

(Bottiglieri et al., 2000). Since the idea that folate deficiency might cause DNA damages was raised (Blount et al., 1997), the possible interaction of folate deficiency and carcinogenesis has been discussed, focusing on breast cancer (Zhang et al., 2005), ovarian cancer (Kotsopoulos, Hecht, Marotti, Kelemen, & Tworoger, 2010), prostate cancer (Collin et al., 2010; Pelucchi et al., 2005), colon tumors (Phelip et al., 2008) and so on. The relationship between anemia and a risk of hip fracture is suggested. Folate administration was reported to reduce the risk of hip fracture in the elderly (Sato, Honda, Iwamoto, Kanoko, & Satoh, 2005). Another big issue has been discussed in the literature that low serum folate levels increase serum homocysteine levels and that elevated homocysteine is associated with a high risk of cardiovascular disease due to its atherogenic action. Many trials for lowering homocysteine levels to prevent primary and secondary cardiovascular disease have been reported (Saposnik et al., 2009; SEARCH Collaborative Group, 2010; Toole et al., 2004; Wald et al., 2011). A unique project in which taking of 400 μg folic acid per day is recommended for community-dwelling people has been carried out in Sakado City, Saitama since 2006. This project has been advanced by Prof. Yasuo Kagawa (Kagawa Nutrition University) and successfully resulted in tremendous reduction of medical expenses by preventing cardiovascular diseases, fractures and so on (Hiraoka et al., 2009).

Infectious diseases frequently occur on respiratory and urinary tracts among the elderly. Immune dysfunction is known to be an age-related pathological state and evoked by micronutrient deficiencies. Lymphocyte-mediated immunity is reported to be decreased in folate deficient humans (Dhur, Galan, & Hercberg, 1991), and to be improved by dietary folate in rats (Field, Van Aerde, Drager, Goruk, & Basu, 2006). Figure 4 shows the changes in the incidence of fever events in our geriatric facility during a two years and nine month period. Patients were divided into groups according to their basal values of serum folate, low folate and high folate groups. The incidence of fever events was decreased in the high folate group after the folic acid-supplementation. This is thought to be another benefit of folic acid-fortification on immunity in addition to above mentioned effects of folic acid on many pathologies. It will be hoped that folic acid-fortified foods come into much wide use in Japan.

Conclusion

Folate deficiency was found in 38% of the patients in our geriatric facility. Taking folic acid-fortified rice effectively corrected the folate deficiency. Concomitantly, macrocytic status of red blood cells was turned to be near normocytic, and fever incidence among patients was decreased. This is the first report that reveals the nutritional benefits of folic acid-supplementation to geriatric patients.

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