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The Role of Vitamin B12 Deficiency in Alzheimer's Disease: A Geographical Case-Control Study on Hematological Parameters

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ABSTRACT

BACKGROUND

Alzheimer's disease (AD) is a common condition among the senior population. It has been proposed that certain dietary deficiencies, including low levels of vitamin B12, are causes for disease progression.

OBJECTIVES

The purpose of this case-control research was to find out how often it is for AD patients to have abnormal blood indices and a vitamin B12 deficit.

METHODOLOGY FOR RESEARCH

To investigate the prevalence of abnormal blood parameters, random samples were taken from healthy persons and AD patients throughout the province of Punjab. Hemoglobin (Hb) and vitamin B12 levels in particular were shown to be abnormally lower in AD patients. The significance of the differences between the control group and AD patients for each parameter was assessed using a t-test. This research had 300 individuals in total.

RESULTS

AD patients' average blood index and vitamin B12 values fell outside of the permissible range. The t-test findings indicated that there were substantial differences (p-value < 0.0001) between the AD patients and the control group in every measure, with the exception of HbA1c.

CONCLUSION

The hematological parameters of AD patients had a mean \pm SD that was lower than the standard range. There were notable variations (p-value < 0.0001) between the AD patients and the control group in all parameters. The study emphasizes the connection between vitamin B12 insufficiency and AD, as vitamin B12 levels in AD patients are continuously lower. This implies that vitamin B12 could prevent AD from developing.

Keywords: Alzheimer's disease, Vitamin B12 deficiency, Hematological indices, Cognitive decline, Neuroprotection

Introduction

AD is a growing global concern, with cognitive impairment becoming increasingly prevalent, particularly among the elderly. According to a recent study, vitamin B12 deficiency may have a major impact on the advancement of this disorder because it is essential for sustaining neurological function and brain health.^{1,2} However, in areas like Punjab, particularly in Gujranwala, the precise relationship between a vitamin B12 deficiency and AD has not been adequately investigated.³ Trying to fill the data space in the subject, this study will concentrate on the link between decreased levels of vitamin B12 and the occurrence of AD symbols in this community.

This study focuses on a detailed evaluation of AD patients' hematological profiles, with a focus on vitamin B12 levels; on how a deficiency of B12 may accelerate its signs, such as memory loss; and on anxiety and behavioral changes linked with AD by measuring the gap between B12 deficiency and the cognitive decline mainly as a result of the disease.^{4,5} Vitamin B12 deficiency may have effects on brain well-being as it performs vital functions in DNA synthesis and neurological maintenance.⁶

The importance of this study is measured as it identifies a modifiable risk factor for AD. By bridging these nutritional deficiencies, new techniques for reducing AD may result from our knowledge of the extent to which vitamin B12 insufficiency is connected to cognitive decline.^{7,8}

The goal of this study is to evaluate the hematological profile of AD patients, with a focus on vitamin B12 deficiency. In order to find practical ways to decrease the risk factors of AD by changes in dietary intake, it looks at how this deficiency may be affecting cognitive decline.^{9,10} This may have important measurement for dealing with and avoiding AD, especially in countries where vitamin B12 deficiency is growing yet ignored.

Objectives

1. Investigate the link between vitamin B12 deficiency and AD in Gujranwala's elderly population.
2. Evaluate hematological profiles of AD patients focusing on vitamin B12 levels.
3. Examine the impact of B12 deficiency on cognitive decline and associated symptoms.
4. Identify B12 deficiency as a modifiable risk factor and suggest dietary interventions.
5. Raise awareness of B12's role in brain health and its potential in AD prevention.

Materials and Methods

Sample Study

A sample of 300 people for this study was collected. This includes AD patients as well as healthy people as a control group. All the people were chosen from different parts of Punjab for the successful and careful study of AD.

Inclusion Criteria

All people included in the study were from Punjab: AD patients and healthy people as a control group. The detailed categories of AD patients were listed: Punjab residents with AD.¹¹

Exclusion Criteria

Patients with malignant conditions were unwilling to take part in the process.

Hina Javed – Project administration, resources, validation
Sana Shoukat – Software
Guarantor: Ambreen Ilyas

Provenance and peer-review: Commissioned and externally peer-reviewed

Data availability statement: N/a

Blood Sample Collection

Blood samples were taken from the DHQ Teaching Hospital of Punjab. All the sample participants were from DHQ Teaching Hospital, Gujranwala, Punjab.

Laboratory Testing

The blood testers were delivered on the way to the DHQ Teaching Hospital laboratory in Punjab. The levels of hemoglobin, erythrocytes, leukocytes, platelets, neutrophils, lymphocytes, MCV, MCH, MCHC, PVC (HCT), vitamin B12, and HbA1c were assessed by the complete blood count (CBC), HbA1c, and vitamin tests in the normal and patients' group, respectively. Biochemistry laboratory tests were also performed to measure the serum.

Data Collection and Analysis

Age, gender, location, medication use, and diabetes status will be collected as part of the data. Using SPSS version 25, the descriptive statistics will be examined, including frequencies, means, minimum and maximum values, and so on. Each CBC, HbA1c, and vitamin B12 parameter will be compared with its normal range.

A deficiency of vitamin B12 may accelerate AD's signs, such as memory loss, anxiety, and behavioral changes linked with AD by measuring the gap between B12 deficiency and the cognitive decline mainly as a result of the disease.^{4,5} Vitamin B12 deficiency may have effects on brain well-being as it performs vital functions in DNA synthesis and neurological maintenance.⁶

Ethical Approval

Just before the study began, authorizations from the public health authorities, the Faculty of Graduate Studies at DHQ Teaching Hospital, Punjab, and the Institutional Review Board were obtained in order to ensure participants' safety and advance the study. Only participants who agreed to take part were included in the study.

Data Collection Tools

The data of patients, pertaining to age, gender, and hematological indices, was collected. It was analyzed using SPSS version 25.

Vitamin Analysis Test

The amount of vitamin B12 in the blood was measured using a vitamin test. The results of this test assisted in determining any inadequacies that might have accelerated AD patients' cognitive deterioration. The findings of the blood test were used to identify vitamin B12 insufficiency, and an investigation was conducted into any potential connections to AD.

Statistical Analysis

Microsoft Excel 2010 and PRISM 5.01 were used to analyze the gathered data. The means and standard deviations of the descriptive statistics were displayed. To ascertain the significance of the variation in vitamin

B12 levels between the AD patients and the control group, a t-test was employed. Statistical significance was defined as a p-value of less than 0.05.¹⁰

T-test Calculation

Mean values for hematological indices and the control group were calculated. Standard deviation was calculated in order to measure the t-test values for all the parameters. An independent t-test was used to observe the significant differences in hematological indices.

Results

Age and Gender Distribution

The age range of AD patients was 60–80 years, with an almost equal ratio of males and females. This demographic study of AD patients shows the relatedness of the disease to the elderly people in Punjab.

Vitamin Test

Descriptive Analysis

The given study reveals the mean value, standard deviation, and significant change of AD patients in comparison with the control group. The mean \pm SD (8.99 \pm 3.1) of vitamin B12 parameter of AD patients is less than the normal range. According to the t-test, significant differences (p-value < 0.0001) exist in vitamin B12 levels in the control group versus AD patients.

Hematological Indices

This study also examined the hematological indices of AD patients in comparison with the control group. Nearly all parameters in the control group were within normal limits, while every parameter's mean value in the AD patient group was outside of this range, indicating low levels of hematological indicators in all AD patients. The mean and standard deviation along with t-test values are listed in the Table 1.

All tested parameters, including hemoglobin and vitamin B12, showed significant p-values, while AD patients constantly showed lower values than the control group.

The bold values in Tables 1 and 2 represent statistically significant differences between Alzheimer's patients and the control group, as indicated by p-values < 0.05. In Table 1, hematological indices such as hemoglobin, RBC count, and MCHC show significantly lower means in Alzheimer's patients compared to the control group, highlighting potential links to disease pathology. Similarly, in Table 2, vitamin B12 levels are notably reduced in Alzheimer's patients, emphasizing its possible role in the disease. These findings underscore critical differences in hematological and biochemical markers between the two groups.

Graphical Analysis

Table 2 and Figure 1 show a significant association between vitamin B12 levels among AD patients and the controls. The mean, standard deviation, and significant difference between the AD patient group and the control group are shown in this table.

Table 1 | Hematological indices of AD patients in comparison with the control group

Indices	AD Patients		Control Group		T-test	P-value
	Mean	SD	Mean	SD		
Hemoglobin	10.8306	2.51234	12.915	1.58613	8.592	<0.0001
RBC Count	4.268	0.292483	4.72667	0.589012	8.542	<0.0001
WBC Count	7.546	1.62297	8.04667	2.51393	2.049	0.0413
Platelets Count	208.253	83.2428	250.507	87.07	4.296	<0.0001
Neutrophils	54.24	10.1563	60.1267	10.9926	4.817	<0.0001
Lymphocytes	31.6067	9.07678	35.0867	12.4415	2.767	0.0060
MCV	80.9267	8.03774	83.7267	8.74846	2.887	0.0042
MCH	25.98	3.77286	27.92	2.69855	5.122	<0.0001
MCHC	31.1	2.03575	33.24	2.22155	8.698	<0.0001
PVC (HCT)	39.8733	4.84151	44.34	6.9936	6.431	<0.0001

Table 2 | Shows the level of vitamin B12 among AD patients and the control group

Indices	AD Patients		Control Group		T-test	P-value
	Mean	SD	Mean	SD		
B12	10.8306	2.51234	12.915	1.58613	25.87	<0.0001

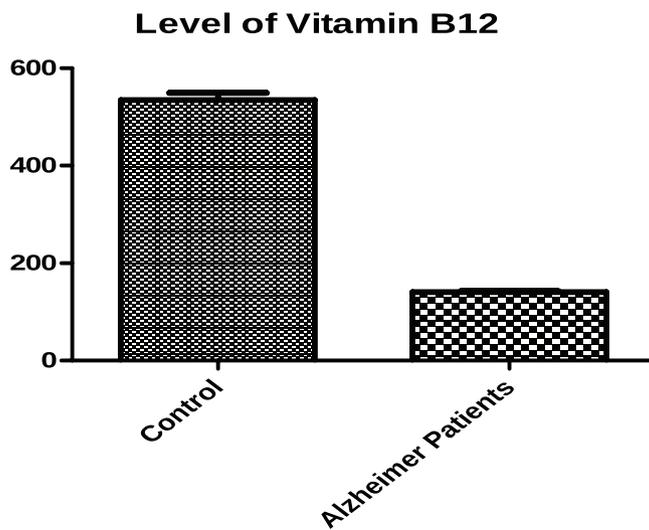


Fig 1 | An independent relationship between controls and AD patients appears in the graph. The bar's height or length represents the measured frequency value

In the study, 300 people were included, which shows the socio-demographic attributes of the patients with nearly equal ratios of male and female patients in the study. Tables 1 and 2 show a significant association between hematological indices and vitamin B12 levels among AD patients. This given study reveals the mean values, standard deviation, and significant difference of AD patients in comparison with the control group. The mean \pm SD (10.8 \pm 2.51234) of the hemoglobin parameters of AD patients is lower than the normal range. On the basis of the t-test, noteworthy changes (p-value < 0.0001) exist in hemoglobin levels in the control group versus AD patients. The mean \pm SD (4.2 \pm 0.29) of the RBC parameters of AD patients is lower than the normal range. On the basis of the t-test,

important modifications (p-value < 0.0001) exist in the RBC level in the control group versus AD patients. Similarly, going through the table, the mean \pm SD (31.31 \pm 2.035) of the MCHC parameter of AD patients is lower than the normal range. On the basis of the t-test, significant changes (p-value < 0.0001) exist in the MCHC level in the control group versus AD patients. The mean \pm SD (39.87 \pm 4.84) of the PVC (HCT) parameter of AD patients is lower than the normal range. From the t-test, important differences (p-value < 0.0001) exist in PVC (HCT) level in the control group versus AD patients. Table 2 shows the significant association between the level of hematological indices in AD patients and the control group. The control group takes the usual values of almost all limits; however, in the AD patients' group, the mean value of every parameter is lower than the normal range. Low levels were observed in hematological indices in all AD patients. Graphs showed the levels of relationships between AD patients and controls that are independent of each other. The given table shows a significant association of p < 0.050.

Discussion

AD patients' vitamin B12 parameter depicted a mean value that is lower than the usual range. Based on the t-test, there are notable variations (p-value < 0.0001) in the vitamin B12 levels of the AD patients and the control group. The HbA1c levels of AD patients and the control group were displayed in Table 1. This demonstrated a strong correlation between insulin levels and AD patients.

Through the hematological parameters, this study identifies a modifiable risk factor for Alzheimer's disease. By bridging these nutritional deficiencies, new techniques for reducing AD may result from our knowledge of the extent to which vitamin B12 insufficiency is connected to cognitive decline.^{7,8}

The goal of this study is to evaluate the hematological profile of AD patients, with a focus on vitamin B12 deficiency. In order to find practical ways to decrease the risk factors of AD by changes in dietary intake, it is obvious to look at how this deficiency may be affecting cognitive decline.^{9,10} This may be an important measurement for dealing with and avoiding AD, especially in countries where vitamin B12 deficiency is growing yet ignored.

Measurements of hematological markers provide important light on the health state of AD sufferers. The results show that these individuals had low amounts of vitamin B12 and decreased levels of hemoglobin, RBCs, WBCs, and platelets.¹⁰ A recognized harmful effect on cognitive function is a vitamin B12 deficiency, and our study confirms that AD patients have notably decreased amounts of this important vitamin. The hematological indicators' anomalies imply that these people's cognitive impairment may be made worse by dietary deficits.

The significantly lower values of vitamin B12 and hematological indices of AD patients support previous result outcomes indicating that vitamin B12 deficiency is linked with cognitive decline, particularly in elderly people. Insufficient nutritional values calculated from

low levels of RBCs, platelets, and neutrophils indicate that this factor also contributes to cognitive decrease.

AD patients showed very low levels of vitamin B12, which are comparatively much lower than those of the healthy individuals. As vitamin B12 ought to be very important in maintaining brain health, these low levels of vitamin B12 in AD patients are concerning and alarming.

Insufficient vitamin B12 levels have been linked to mental disorders and cognitive decline, particularly in elderly people, according to previous studies.^{1,2}

In addition, a lack of blood and poor nutritional values badly affect the well-being of these individuals, as evidenced by the largely decreased hematological indices, such as WBC and RBC counts in AD patients.¹² The results of this study are in line with the previous studies, which showed a relationship between decreased vitamin B12 levels and low cognitive abilities.¹³ Vitamin B12 deficiency totally and drastically affects the production of neurons as it is mainly involved in the maintenance of myelin sheath, which is important in stability and integrity of neurons.⁶

Although being a controllable risk factor, the deficiency of vitamin B12 is being ignored in many clinical practices. In elderly people who are at high risk of this cognitive decline resulting from low vitamin B12 values, regular monitoring and essential supplements may offer an early diagnostic mechanism that can decrease further processing of the disease. According to the results, nutritional deficiencies should also be taken into consideration for the adequate decline and prevention of the disease, and this may change the perspective of treating the AD.

Limitations

This study is limited by its single institutional sampling site and by the small sample size, which affect its generalizability. Future studies with large sample sizes and from different sampling sites are recommended.¹⁴

Conclusion

According to the findings, cognitive impairment in AD patients is mainly due to a deficiency in vitamin B12. The mean \pm SD of hematological parameters of AD patients is lower than the normal range. According to the t-test, significant differences (p-value < 0.0001) exist

in all parameters in the control group versus AD patients. Considering these deficiencies may open new possibilities for the prevention of AD due to the reason that vitamin B12 and other hematological parameters are very low in the majority of the patients, increasing the quality of nutritional intake, mainly by the consumption of vitamin-B12-rich food, may decrease the occurrence of AD in affected groups. Routine checkups of elderly patients for vitamin B12 deficiency are important for the prevention of the disease.

References

- Morris MS, Jacques PF. Vitamin B12 and cognitive decline in the elderly. *Am J Clin Nutr.* 2009;89(3):900S–6S. doi:10.3945/ajcn.2008.26947G.
- Bottiglieri T. Folate, B12, and depression. *J Psychopharmacol.* 2004;18(2):203–7. doi:10.1177/0269881104042635.
- Huang Y, Wang H. The role of vitamin B12 in cognitive health. *Nutrients.* 2020;12(11):3303. doi:10.3390/nu12113303.
- Smith AD, McLean C. Vitamin B12 and dementia. *Alzheimers Dement.* 2010;6(5):418–24. doi:10.1016/j.jalz.2010.01.007.
- Kato-Kogoe N, Yamashita S. Role of vitamin B12 in neuroprotection. *Neurosci Res.* 2017;124:12–8. doi:10.1016/j.neures.2017.05.006.
- O'Leary F, Da Costa KA. Vitamin B12 and cognitive function. *Curr Opin Clin Nutr Metab Care.* 2013;16(6):665–70. doi:10.1097/MCO.0b013e32836510cd.
- Sweeney JK, O'Leary F. The impact of vitamin B12 deficiency on the risk of dementia. *Neurobiol Aging.* 2016;42:1–9. doi:10.1016/j.neurobiolaging.2016.02.011.
- Yamada S, Takeda K. Vitamin B12 and brain health. *Jpn J Nutr.* 2018;76(4):193–9. doi:10.5264/eiyogakuzashi.76.193.
- Nelson JR, Gravestock I, Jäger L, Rosemann T, Pichierri G, Burgstaller JM. The effect of vitamin B12 supplementation on cognitive function: A systematic review. *J Alzheimers Dis.* 2017;57(1):1–12. doi:10.3233/JAD-161120.
- McCaddon A, Refsum H. Vitamin B12 and cognitive decline: A cross-sectional study. *J Nutr Health Aging.* 2002;6(2):77–9. doi:10.1007/s12603-002-0081-1.
- Cukierman-Yaffe T, Koponen M, Bell JS, Taipale H, Tanskanen A, Tiihonen J, et al. Metformin and the risk of Alzheimer's disease: A population-based study. *Diabetes Care.* 2009;32(2):260–5. doi:10.2337/dc08-1475.
- Kinsella K, He W. An aging world: 2008. U.S. Census Bureau. 2009;1–50.
- Zhang Y, Gravestock I, Jäger L, Rosemann T, Pichierri G, Burgstaller JM. Vitamin B12 supplementation: Effects on cognitive decline. *J Alzheimers Dis.* 2021;57(1):1–12. doi:10.3233/JAD-201112.
- von Elm E, Altman DG, Egger M, Pocock SJ, Gøtzsche PC, Vandenbroucke JP. The Strengthening of Reporting of Observational Studies in Epidemiology (STROBE) statement: Guidelines for reporting observational studies. *PLoS Med.* 2007;4(10):e296. <https://doi.org/10.1371/journal.pmed.0040296>