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FORMULATION AND EVALUATION OF VITAMIN B12 NASAL SPRAY FOR ENHANCED PATIENT COMPLIANCE

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ABSTRACT
Vitamin B12 is an essential micronutrient required for DNA synthesis, neurological function, and red blood cell maturation. Deficiency of Vitamin B12 is a global health concern, leading to anaemia, neuropathy, and neurocognitive impairment. Conventional therapy involves oral supplementation and intramuscular injections, but these routes face significant challenges. Oral absorption is limited by gastrointestinal factors, while parenteral injections are painful, inconvenient, and associated with poor patient compliance. Therefore, alternative delivery approaches are urgently needed. The nasal route of administration is an attractive option due to its large surface area, high vascularization, and avoidance of hepatic first-pass metabolism. Nasal sprays are patient-friendly, non-invasive, and capable of delivering therapeutically effective drug concentrations. In this study, a Vitamin B12 nasal spray formulation was developed using solubilizers, stabilizers, and absorption enhancers. The formulation was subjected to extensive evaluation, including physicochemical properties, spray performance, drug content, in-vitro diffusion, and stability studies. The optimized spray demonstrated a pH compatible with the nasal cavity, low viscosity for easy actuation, and uniform drug content. Spray characterization revealed consistent droplet size and spray angle, while in-vitro diffusion studies confirmed enhanced permeation compared to plain Vitamin B12 solution. Stability studies over three months under ICH accelerated conditions demonstrated retention of drug content, clarity, and performance. This study highlights the potential of Vitamin B12 nasal spray as a practical alternative to oral and injectable dosage forms. Further in-vivo studies and clinical trials are warranted to confirm its therapeutic effectiveness and potential commercialization.

KEYWORDS: Vitamin B12, Nasal spray, Intranasal drug delivery, Bioavailability, Patient compliance.

INTRODUCTION
Vitamin B12, also known as cobalamin, is an essential water-soluble vitamin that plays a vital role in various biochemical and physiological processes. It is a cofactor in DNA synthesis, fatty acid metabolism, and myelin sheath formation, making it indispensable for normal haematological and neurological functions. Deficiency of Vitamin B12 is a common global health issue, particularly among the elderly, vegetarians, vegans, and patients with gastrointestinal malabsorption syndromes such as pernicious anaemia, Crohn's disease, or after bariatric surgery. Clinical manifestations of deficiency include megaloblastic anaemia, fatigue, cognitive decline, neuropathy, and in severe cases, irreversible neurological damage.[1]

The prevalence of Vitamin B12 deficiency is estimated to range from 6–20% in adults under 60 years, and up to 30–40% in older adults, depending on dietary habits and health conditions. In India and other developing nations, the deficiency is particularly high due to vegetarian diets and lack of fortified foods. Thus, ensuring adequate Vitamin B12 supplementation is an important public health priority.[1]

Traditionally, Vitamin B12 supplementation has been provided via oral tablets or intramuscular injections. Oral therapy is often ineffective in patients with impaired absorption, since it requires intrinsic factor-mediated uptake in the ileum. Furthermore, oral bioavailability is highly variable and typically less than 2%. On the other hand, intramuscular injections bypass absorption barriers and ensure adequate serum levels, but they are invasive, painful, require trained personnel, and lead to poor long-term compliance. Repeated injections also increase the risk of injection-site reactions, infections, and needle-associated anxiety in patients.[2]

To overcome these limitations, alternative drug delivery systems have been investigated, including sublingual tablets, buccal films, transdermal patches, and intranasal sprays. Among these, the nasal route has gained

significant attention in recent years as a promising approach for systemic drug delivery. The nasal cavity is highly vascularized, offering a large absorptive surface area (~150 cm²), and allows direct entry of drugs into systemic circulation, bypassing gastrointestinal degradation and hepatic first-pass metabolism. Moreover, nasal sprays are non-invasive, self-administered, and portable, which enhances patient adherence compared to injections.[3]

The formulation of Vitamin B12 as a nasal spray offers several clinical advantages.

1. Rapid absorption and onset of action due to the rich blood supply in the nasal mucosa.
2. Improved patient compliance, especially in elderly patients and those requiring long-term therapy.
3. Avoidance of needles, eliminating injection-related pain and anxiety.
4. Bypassing gastrointestinal tract and first-pass metabolism, which improves systemic bioavailability.
5. Potential for direct nose-to-brain delivery, which may have therapeutic benefits in neurological complications of Vitamin B12 deficiency.

Previous research has reported promising outcomes with intranasal delivery of Vitamin B12, showing plasma concentrations comparable to intramuscular injections. However, the success of a nasal formulation depends on critical factors such as drug solubility, stability, permeation through the nasal mucosa, and spray device performance. Formulation excipients, such as solubilizers, stabilizers, mucoadhesive agents, and absorption enhancers, play a crucial role in optimizing these parameters.[4,5]

Despite encouraging studies, there remains a need for well-optimized, stable, and reproducible Vitamin B12 nasal spray formulations that can be developed at a scale suitable for clinical use. Thus, the present study was undertaken to formulate and evaluate a Vitamin B12 nasal spray with the following objectives.

- To develop a stable and patient-friendly nasal spray formulation containing Vitamin B12.
- To characterize its physicochemical and performance properties, including pH, viscosity, clarity, and drug content.
- To assess in-vitro diffusion performance across nasal mucosa.
- To evaluate stability under accelerated storage conditions as per ICH guidelines.

The outcomes of this work are expected to contribute towards the development of a convenient, non-invasive, and effective alternative to traditional oral and injectable Vitamin B12 supplementation, ultimately improving patient adherence and therapeutic outcomes.

MATERIALS AND METHODS

Chemicals and Reagents

Drug: Cyanocobalamin (Vitamin B12) was obtained from a certified pharmaceutical supplier with a purity of >99%.

Excipients.

Buffer system: Phosphate buffer saline (PBS) pH 5.5–6.0 (to mimic nasal fluid environment).

Solubilizer: Polyethylene glycol 400 (enhances solubility of Vitamin B12).

Stabilizer: Glycerine (maintains isotonicity and prevents degradation).

Preservative: Benzalkonium chloride (0.01% w/v, prevents microbial growth).

Absorption enhancer: Chitosan (0.25% w/v, improves mucosal adhesion and permeation).

Membrane for diffusion studies: Freshly excised goat nasal mucosa, obtained from a local slaughterhouse and used within 2 hours.

Instruments: Digital pH meter, Brookfield viscometer, Franz diffusion cell (effective diffusion area ~2 cm²), UV–VIS spectrophotometer (λmax = 361 nm for Vitamin B12).

Formulation of Nasal Spray

1. Preparation of buffer: Phosphate buffer was prepared and adjusted to pH 5.8 using NaOH/HCl.
2. Dissolution of drug: Accurately weighed Vitamin B12 was dissolved in the buffer under magnetic stirring.
3. Addition of excipients: PEG-400 and glycerine were added slowly with continuous stirring.
4. Enhancer incorporation: Chitosan was dissolved separately in 1% acetic acid and added to the solution.
5. Sterilization: The solution was filtered through a 0.22 µm sterile filter membrane.
6. Filling and packaging: The sterile solution was transferred aseptically into pre-sterilized amber glass nasal spray bottles fitted with metered-dose pumps (100 µL per actuation).

Evaluation of Formulation

1) Physicochemical Properties

- pH: Measured using calibrated pH meter (triplicate readings).
- Viscosity: Determined using Brookfield viscometer at 25 °C.
- Clarity: Checked visually against black and white backgrounds under good illumination.
- Drug content uniformity: 1 mL sample diluted appropriately and analysed spectrophotometrically at 361 nm.

2) Spray Performance

- Spray angle: Determined by spraying onto Whatman filter paper placed at 5 cm distance and measuring diameter.
- Droplet size: Evaluated using laser diffraction technique (Malvern particle sizer).

- Actuation volume: Each spray was weighed, and mean volume per actuation was calculated.

3) In-vitro Diffusion Study

- Setup: Franz diffusion cell with goat nasal mucosa clamped between donor and receptor chambers.
- Donor chamber: Contained 1 mL of nasal spray formulation.
- Receptor chamber: Contained PBS (pH 6.4) maintained at 37 ± 0.5 °C with constant stirring.
- Sampling: Aliquots withdrawn at 10, 20, 30, 45, 60 minutes and replaced with fresh buffer.
- Analysis: UV–VIS spectrophotometer at 361 nm.
- Parameters calculated: Cumulative % drug release, diffusion rate, and flux.

4) Stability Study

Conducted as per ICH Q1A (R2) guidelines:

- Accelerated condition: 40 °C ± 2 °C / 75% ± 5% RH.
- Duration: 3 months.
- Sampling intervals: 0, 1, and 3 months.
- Parameters monitored: pH, viscosity, drug content, clarity, and spray performance.

5) Microbial Testing

- Sterility test: Performed as per IP guidelines by inoculating samples in fluid thioglycolate medium and soybean-casein digest medium, incubated for 14 days.
- Acceptance criteria: No growth should be observed.

RESULT AND DISCUSSION

The Vitamin B12 nasal spray was successfully formulated and evaluated. The results of physicochemical properties, spray performance, in-vitro diffusion, and stability studies are presented in the following tables and figures.

Table 1: Physicochemical Properties of Vitamin B12 Nasal Spray.

Parameter	Optimized Formulation	Standard Requirement
pH	5.8 ± 0.1	5.0 – 6.5
Viscosity (cP)	28 ± 2	< 50
Clarity	Clear, no turbidity	Should be clear
Drug Content (%)	98.5 ± 1.2	95 – 105%

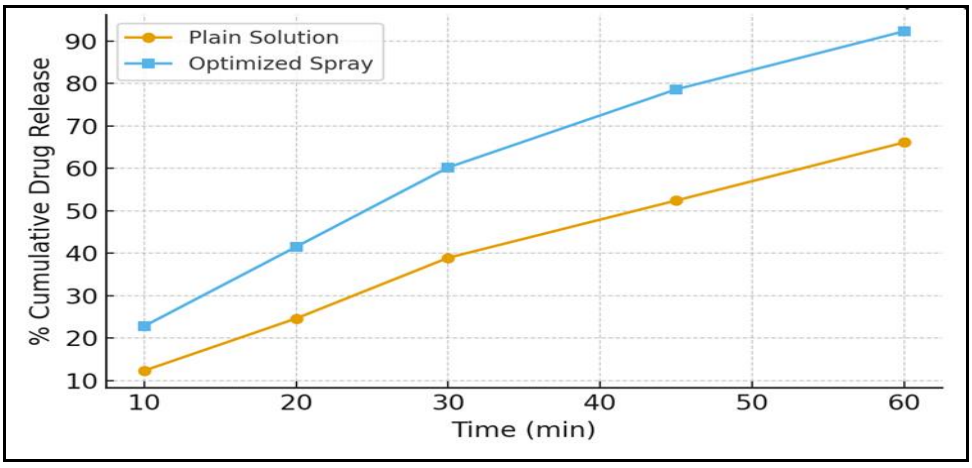

Fig. 1: In-Vitro Diffusion Profile of Vitamin B12 Nasal Spray.

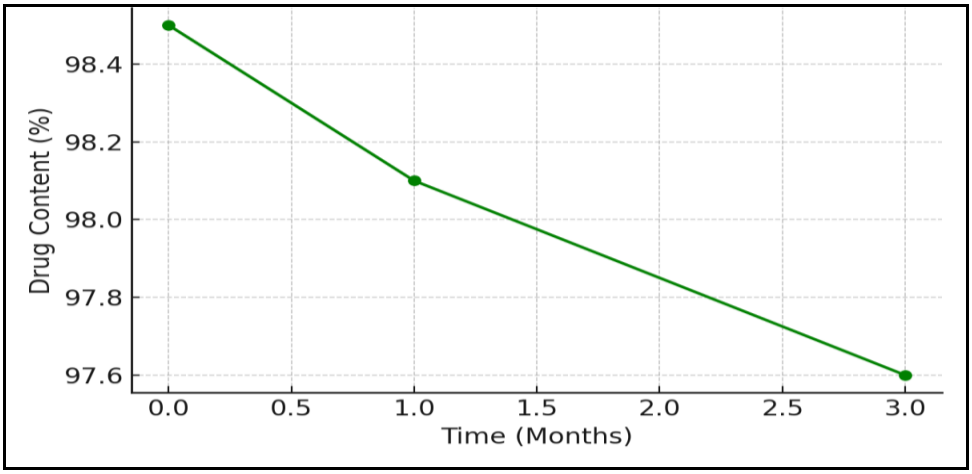

Fig. 2: Stability Profile of Vitamin B12 Nasal Spray.

The results indicated that the nasal spray formulation had suitable physicochemical characteristics. The pH was within the acceptable nasal range (5.0–6.5), ensuring minimal irritation upon administration. The viscosity was low, allowing effective atomization during actuation. Drug content analysis confirmed uniformity across the formulation. The in-vitro diffusion profile demonstrated significantly improved permeation with the optimized spray compared to plain solution, highlighting the role of chitosan as a permeation enhancer. Stability studies under accelerated conditions confirmed that the formulation retained its clarity, viscosity, and potency for three months. These findings support the feasibility of Vitamin B12 nasal spray as an alternative to conventional therapies.

CONCLUSION

The present study successfully developed and evaluated a Vitamin B12 nasal spray with favourable physicochemical, performance, and stability characteristics. The optimized formulation demonstrated enhanced drug permeation compared to plain solution, indicating improved bioavailability. Nasal delivery offers a patient-friendly, non-invasive alternative to oral and injectable Vitamin B12 supplementation, with the potential to improve compliance and therapeutic outcomes. Future work should focus on in-vivo pharmacokinetic studies and clinical trials to establish efficacy and support potential commercialization.

REFERENCES

1. Shipkova M, et al. Vitamin B12 deficiency – modern approaches to diagnosis and therapy. Clin Chim Acta, 2019; 495: 39–45.
2. Allen LV, Popovich NG, Ansel HC. Pharmaceutical Dosage Forms and Drug Delivery Systems. 10th ed. Wolters Kluwer: 2021.
3. Illum L. Nasal drug delivery – possibilities, problems and solutions. J Control Release, 2003; 87(1–3): 187–198.
4. Sadoff J, Le Gars M, Shukarev G, et al. Intranasal drug delivery: Current and future prospects. Drug Delivery and Translational Research, 2020; 10(4): 1–10.
5. Patel A, et al. Advances in nasal drug delivery systems: A review. Int J Pharm Sci Res, 2019; 10(2): 529–540.
6. Hussain A, et al. Nasal route of drug delivery: Recent advances and future prospects. Int J Pharm Investing, 2021; 11(3): 188–195.