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Evaluation of New Strength Formulation of Anti-Cancer Drug and its Characterization

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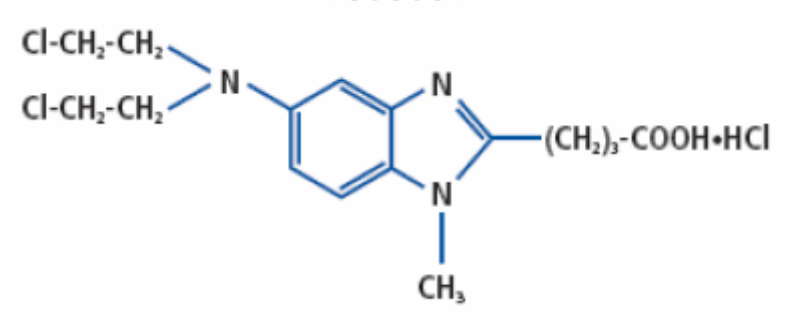
ABSTRACT:

B-cell chronic lymphocytic leukemia (B-CLL), also known as chronic lymphoid leukemia (CLL), is the most common type of leukemia. Leukemias are cancers of the white blood cells (leukocytes). Bendamustine (INN, trade names Ribomustin, Treanda, Bendeka and Belrapzo) is nitrogen mustard used in the treatment of chronic lymphocytic leukemia and lymphomas. It belongs to the family of drugs called alkylating agents. Bendamustine Hydrochloride is commercially available in the market as a lyophilized dosage form across the globe whereas in US, non-aqueous solution form is also commercially available. Package inserts evaluation of all available 3 products and also evaluating various articles; it is learnt that there is a scope to evaluate novel strength requirement for the effective use and hence attempt is made to present novel intermediate strength-based formulation. Also enough literature is available that Bendamustine Hydrochloride is very unstable in the liquid dosage form. Hence an attempt for developing a simple Bendamustine Hydrochloride formulations is made. The data suggested that there is a need to revisit the formulation by exploring new solvents.

INTRODUCTION:

Cancer is a disease of uncontrolled cell division, invasion and metastasis[1]. It is generally considered to be due to the clonal expansion of a single neoplastic cell. Cancers are classified in two ways[2]: by the type of tissue in which the cancer originates (histological type) and by primary site, or the location in the body where cancer first developed. It is also being studied for the treatment of sarcoma[3]. Bendamustine was first synthesized in 1963 by Ozegowski and Krebs in East Germany (the former German Democratic Republic). It undergoes hydrolytic degradation in the presence of water[4]. It is a white, water-soluble microcrystalline powder with amphoteric properties[5]. Until 1990 it was available only in East Germany. East German investigators found that it was useful for treating chronic lymphocytic leukemia, Hodgkin'sdisease, non-Hodgkin's lymphoma, multiple myeloma and lung cancer[6]. The IUPAC name of Bendamustine Hydrochloride is lH-benzimidazole-2-butanoic acid,5-[bis(2-chloroethyl)amino]-lmethyl-,monohydrochloride. Its empirical molecular formula is $C_{16}H_{21}Cl_2N_3O_2$.HC1, and the molecular weight is 394.7. Bendamustine hydrochloride contains a mechlorethamine group and a benzimidazole heterocyclic ring with a butyric acid substituent, and has the following structural formula[7].



In India, the drug Bendamustine Hydrochloride was approved as the Lyophilized powder Injection 100mg/ vial for the treatment of patients with chronic lymphocytic leukemia. As per the literature available, the lengthy exposure of Bendamustine to water during the reconstitution process increases the potential loss of potency and impurity formation due to the hydrolysis of the product by water. Lyophilization is a time-consuming, tedious and involves cumbersome procedures. Further, it involves expensive technology to develop a lyophilized product. One of the main disadvantages of lyophilization is the expensive and the lyophilization cycle development and criticality of the freeze-drying process, which requires very low temperatures, can be quite costly. Further, the product needs to be handled with the precautions while dispensing, manufacturing and lyophilization. The lyophilization cycle

recipe needs to be set carefully based on the load of the vials that goes into the lyophilizer. Hence, an attempt to develop a non-lyophilized drug product such as liquid formulation which would offer convenience for practitioners by avoiding the reconstitution step when preparing the drug for administration.

MATERIALS AND METHODS:

Bendamustine Hydrochloride was procured from Keman Chemicals, Gujarat, DMSO was sourced from Sigma-Aldrich, Edetate Di-Sodium and L-Arginine was received as gift sample from Merck. Soya bean oil, Polysorbate 80 and Polyoxy castor oil were purchased from commercial sources. All required chemicals used were of standard grade.

Preparation of Bendamustine Hydrochloride Formulations

A total of 3 formulations were prepared. The concentration chosen of Bendamustine Hydrochloride was 45 mg/mL based on the solubility. Initially, purified water was taken and drug was added and dissolved. Later on, one by one excipient as per below composition is added. Finally, 100% volume was made using the vehicle per below table. There are no pH adjusting agent used in the formulation.

Table No. 1: Formulation of Bendamustine Hydrochloride Injection

S. No.	Ingredients	BF1	BF2	BF3
1	Bendamustine Hydrochloride	45 mg/mL	45 mg/mL	45 mg/mL
2	Soya bean Oil	200 mg/mL	200 mg/mL	200 mg/mL
3	Polysorbate 80	25 mg/mL	25 mg/mL	25 mg/mL
4	Polyoxyl castor oil 35	--	200 mg/mL	200 mg/mL
5	Dimethyl Sulfoxide	--	--	0.3 mL/mL
6	Edetate Disodium	0.1 mg/mL	0.1 mg/mL	0.1 mg/mL
7	L-Arginine	--	--	100 mg/mL
8	Purified Water	QS to 1 mL	QS to 1 mL	QS to 1 mL

Evaluation of Bendamustine Hydrochloride Formulations

Physical evaluation

***Description*:** This is a physical observation made by individual.

pH: pH was measured using pH meter at about 25°C temperature.

Light Transmission: *All the* formulations were tested for light transmission at 650 nm using UV spectrophotometer.

Chemical Evaluation

Assay: HPLC method was adopted to measure the active drug content from the 3 formulations. The active obtained is expressed as a percent of the labeled amount of Bendamustine Hydrochloride content. The obtained value of drug content is expected to be within limits of 90.0% to 110.0% (General compendia like USP & BP requirement).

Related Substances: % content of known and unknown impurities were determined by HPLC method.

RESULTS AND DISCUSSION:

The results are compiled in the table 2. A clear colourless to light yellow colour solution was observed from BF1 to BF3 formulations. pH of all 3 formulations was observed in the range of 2.5 to 3.0 wherein the formulations don't contain pH adjusting agents. This indicates that the pH of the formulations is independent of drug substances though there is a qualitative and quantitative change among the three formulations. It is also noted from the pH trend that all the three formulations indicated that formulation stability is towards the acidic nature as the drug substance is salt of weak acid which has butyric acid moiety. Light transmission measured for the three formulations found between 95 to 100% indicating the clear transmission of the liquid formulation when the each of the formulations was transmitted through UV spectrophotometer at 650 nm. For the chemical analysis of all the three formulations, it was observed that all the three formulations have shown assay value about 98.0 % indicating the correct input of % content of Bendamustine Hydrochloride vs label claim. It also indicates that the analytical method employed for estimating the % content of Bendamsutine Hydrochloride is correct. From the related substances analysis, it was observed that monohydroxy Bendamustine (impurity A) was observed in all the three formulations in a significant amount and other two known impurities such as dihydroxy Bendamustine (Impurity B) and Dimer Impurity (Impurity C) content are satisfactory. However, the content of the single highest unknown impurity is found high in all three formulations.

Table No. 2: Physical and Chemical Evaluation of Aqueous Bendamustine Hydrochloride Formulations

S. No.	Formulation Codes	Description	pH	LT (in %)	Assay (in %)	Related Substances
1	BF1	@	2.94	96.5	97.1%	Imp A:2.58% Imp B:0.16% Imp C:0.14% Single Highest UNK Imp: 0.32% Total Imp: 3.42%
2	BF2	@	2.92	97.1	95.8%	Imp A:3.61% Imp B:0.18% Imp C:0.13% Single Highest UNK Imp: 0.26% Total Imp: 4.31%
3	BF3	@	2.85	97.4	97.6%	Imp A:1.44% Imp B:0.15% Imp C:0.09% Single Highest UNK Imp: 0.22% Total Imp: 2.08%

@: Description: A clear colorless to light yellow color solution. LT is Light Transmission.

Imp A is Impurity A: Monohydroxy Impurity, Imp B is Impurity B:Dihydroxy Impurity and Imp C is Impurity C: Dimer Impurity.

CONCLUSION:

Overall characterization of all the three formulations concluded that no physical description complications were observed. Analytical results of pH and light transmission test parameters were found satisfactory. Chemical evaluation such as assay test parameter result was observed satisfactory. However, for impurity formation such as monohydroxy bendamustine

was observed in the significant levels indicating the hydrolytic degradation nature of impurity A. Though the soya bean oil quantity in each of the formulation was satisfactory levels, but looks that the quantity of soya bean oil is not able to reduce the % content of Impurity A. However, the other two known impurities such as dihydroxy and dimer impurities result in satisfactory. It is also to be noted that % content of unknown impurities is on higher side in all the three formulations. From the above experiment, it can be concluded that bendamustine hydrochloride needs fine-tuning for a lesser quantity of water so as to arrest the degradation as impurity A is a hydrolytic impurity and is observed in the significant levels in all the formulations which is also not in line with the requirements of ICH Q3 B R (2). However, formulation BF3 has lesser impurity levels when compared to other two BF1 and BF2 formulations indicating that the level of water in the formulation plays an important role. As an alternate, the scope of developing nonaqueous Bendamustine Hydrochloride Injection shall be attempted.

REFERENCES:

1. Tripathi K D, Essentials of Medical Pharmacology, 5thed, Jaypee Brothers Medical Publishers (P) Ltd, NewDelhi;2004:769-782.

2. Rey L, May JC, editor. Freeze drying/Lyophilization of pharmaceutical and biological products. 2nd ed, Marcel Dekker Inc New York; 2004:1-32.

3. Harris NL, Jaffe ES, Diebold J *et al*., "World Health Organization classification of neoplastic diseases of the hematopoietic and lymphoid tissues: report of the Clinical Advisory Committee meeting-Airlie House, Virginia, November 1997". J. Clin. Oncol. 17 (12), 3835–3849, PMID 10577857.

4. US Patent application 20110184036; Eagle Pharmaceuticals; Filed to USPTO on Jan 28, 2011.

5. Bagchi S (August 2007). "Bendamustine for advanced sarcoma", Lancet Oncol, 8 (8), 674. doi:10.1016/S1470-2045, (07), 70225- 5. PMID 17726779.

6. Cephalon press release - Cephalon Receives FDA Approval for TREANDA, a Novel Chemotherapy for Chronic Lymphocytic Leukemia". Retrieved 2008-03-23.

7. TREANDA-Prescribing information. 2010 [cited; Available from: http://www.accessdata.fda.gov/drugsatfda_d ocs/label/2010/022249s005lbl.pdf.