STABILITY AND RHEOLOGICAL PROPERTIES OF NOURIVAN™/ANTIOX® BASE WITH SELECTED SOLVENTS

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Abstract

The basic parameters of the Nourivan™/Antiox® base were analysed. The base was mixed in a 1:1 ratio with water, 96% ethanol, castor oil, glycerol and in a ratio of 1:0.5 with castor oil and Peru balsam. The prepared formulations were subjected to rheological tests immediately after preparation and after 4 months since their preparation. Rheological tests were carried out using a Brookfield digital rheometer. It was found that the support retains its homogeneous form when mixed with water, castor oil (1:0.5), ethanol, Peru balsam, glycerol, both on the day of preparation and after the stability test. In rheological tests of the substrate, the lowest rheological stability was demonstrated for the combination with the Peru balsam. The value of the yield stress of the base with the content of Peru balsam was three times lower after 4 months of storage than marked *ex tempore*. The studies carried out will make it possible to assess the risk of using Nourivan™/Antiox® base for formulation with active substances suspended or dissolved in the described solvents.

Rezumat

Au fost analizați parametri bazei Nourivan™/Antiox®. Baza a fost amestecată într-un raport 1:1 cu apă, 96% etanol, ulei de ricin și glicerol și într-un raport de 1:0,5 cu ulei de ricin și balsam de Peru. Formulările preparate au fost supuse testelor reologice imediat după preparare și după 4 luni, utilizând un reometru digital Brookfield. S-a constatat că baza își păstrează forma omogenă atunci când este amestecată cu apă, ulei de ricin (1:0,5), etanol, balsam de Peru, glicerol, atât în ziua preparării, cât și după determinarea testului de stabilitate. În testele reologice ale substratului, cea mai mică stabilitate a fost demonstrată pentru asocierea cu balsamul de Peru. Valoarea tensiunii bazei cu conținut de balsam de Peru a fost de trei ori mai mică după 4 luni de păstrare decât în momentul obținerii *ex tempore*. Studiile efectuate vor face posibilă evaluarea riscului de utilizare a bazei Nourivan™/Antiox® pentru formulările cu substanțe active suspendate sau dizolvate în solvenții descriși.

Keywords: Nourivan™, Antiox®, rheology, stability, yield stress

Introduction

Nourivan™/Antiox® is a new complex emulsion base oil-in-water (o/w). According to the information provided by the manufacturer, it is a complex preparation, mildly hydrophilic in character, with antioxidant and moisturizing properties [1]. The formulation of Nourivan™/Antiox® is suitable for balanced and dehydrated skin. It can be used as an ideal carrier for active pharmaceutical (API) and dermatological (DCI) components susceptible to oxidation. The antioxidant character of this base results from the use of raw materials in its composition, such as ascorbic acid and α -tocopherol. The content of acidum ascorbicum and tocopherol in the base composition may serve as a protection against oxidation of active substances added to the base. Vitamin C is a natural antioxidant displaying antioxidant, anti-inflammatory, photoprotective properties, and is a known biostimulator of collagen synthesis [2, 3].

Because of its *in vivo* inhibitory action on melanin synthesis, vitamin C is useful as a whitening agent in cosmetics [4, 5].

The topical application of ascorbic acid has been shown to significantly elevate cutaneous levels of this vitamin, and this correlates with protection of the skin from UVB-induced oxidative damage as measured by a decrease in UVB erythema and sunburn cell formation [5, 6]. In the presence of α -tocopherol, important lipid-soluble radical scavenger, after neutralization of free radicals; vitamin C not only regenerates itself, but also regenerates vitamin E, which increases their synergistic antioxidant effect. The presence of α -tocopherol in formulations may also have a stabilizing effect on the photo-degradation of ascorbic acid [7-9]. In order to be effective and safe, the medicine should be of appropriate quality and, from the patient's point of view; it should be free of any problems during use. Available literature data indicate that medicinal substances belonging to class II of the Biopharmaceutical

Classification System (BCS) are the subject of numerous publications [10]. Lack of success or reduction of treatment effects of different dermatological use drugs for may result, *inter alia*, from poor solubility of the medicinal substance. Many factors influence the solubility of a medicinal substance, such as, e.g. lipophilicity, molecular weight, crystallographic form or the solvent type used [11-16]. In order to solve this problem, new compositions of ointment vehicles are being developed or existing ones modified. In pharmacy practice, a pharmacist prepares the dermatological preparation for an individual patient, according to an individual recipe - in order to obtain optimal dispersion in the system; he does not introduce the drug directly into the bases but uses appropriate solvents.

The introduction of an active substance into the base depends on its physicochemical properties, in particular on the solubility in typical polar and non-polar solvents used in the preparation of prescription drugs, or the production of new dermatological drugs. The process of combining active ingredients with the base often requires the use of appropriate auxiliary substances. Obtaining a permanent system during this process and appropriate functional properties involves conducting experiments in order to understand the technological aspects concerning, *inter alia*, the ability to bind water and other hydrophilic or lipophilic solvents of the tested base.

The aim of the study was to assess the physicochemical properties of the NourivanTM/Antiox[®] base after addition of selected solvents used in everyday recipe practice and to assess the stability of the base-solvent combination based on rheological research.

Materials and Methods

Materials. Liquid paraffin (Pharmaceutical Laboratory, Poland), castor oil (Pharma Cosmetic, Poland), rapeseed oil (PPH Galfarm, Poland), 86% glycerol (Pharma Cosmetic, Poland), 96% ethanol, Peru balsam (Pharma Cosmetic, Poland), *Aqua pro usu officinale* (Pharma Cosmetic, Poland), (Avantor Performance Materials, Poland). All materials used in the study were p.a. and satisfy the requirements of standards and certificates. NourivanTM/Antiox[®] was gift from Fagron (Pharma Cosmetic, Poland). This base contains: purified water, cetearyl alcohol, polysorbate 60, C13-C16 isoparaffin, C12-C14 isoparaffin, C13-C15 alkane, glyceryl stearate, PEG-75 stearate, polyacrylate 13, polyisobutate, polysorbate 20, polyurethane-39, stearyl behenate, cetyl alcohol, ascorbic acid, benzoic acid, sodium bisulfite, sorbic acid, tocopheryl acetate.

Water absorption capability tests. The introduction of water into the test base was carried out using an unguator (Unguator[®] ES GAKO) according to FPXI [18]. The addition of water was completed at the moment the test medium acquired the consistency classified into semi-solid dosage forms. The amount

in emulsified water in relation to the medium was calculated from the mass difference after weighing the mortar with the base *ex tempore* and after study completion. The mixing process was carried out at 800 rotations/min at 25°C for 2 minutes. The test was carried out six times, regardless of the method used.

Determination of the degree of mixing of solvents with the tested base. Using Unguator[®], base NourivanTM/Antiox[®] was mixed with selected solvents in a 1:1 ratio with purified water at 22 ± 1°C, 96% ethanol, castor oil, 86% glycerol and in a ratio of 1:0.5 with castor oil and Peruvian balsam. The samples prepared were subjected to rheological study immediately after their preparation and after 16 weeks since the preparation.

Viscosity. The samples were sheared in the space between the rotating cone and the stationary plate of the digital rheometer Brookfield CAP 2000⁺ (Brookfield Engineering Inc.) at temp. 25°C. Within the range of shear rates from 66.66 to 1267 1/s, values of shear stress and viscosity, which correspond to the shear rate used during the experiment, were determined. On the basis of the yield curves, the values of yield stress were determined, using the mathematical Casson model (Rheocalc for Windows). The Casson model is described by the following formula:

$$\sqrt{\sigma} = \sqrt{\sigma_0} + \sqrt{\eta\dot{\gamma}},$$

where: σ = shear stress, σ_0 = yield stress (shear stress at the shear rate aiming at zero), η = plastic viscosity, $\dot{\gamma}$ = shear rate.

Statistical analysis. The experimental results are expressed as mean ± standard deviation (SD). Statistical analysis was carried out using one-way analysis of variance (ANOVA) or Student's t-test. Tukey's post hoc test for multiple comparisons among tested formulations was also used. In order to determine the statistical significance for means with a distribution different from normal or heterogeneous variances, the nonparametric test of Wilcoxon and Kruskal-Wallis was used. The statistical analyses were performed using STATISTICA software (ver. 12). Results at $p < 0.05$, $p < 0.01$ or $p < 0.001$ are denoted by a single (*), double (**) or triple (***) asterisk, respectively. The statistical significance assumed the significance level $p < 0.05$.

Results and Discussion

NourivanTM/Antiox[®] constitutes the base for semi-solid forms of medicines in some countries in Europe and the USA. In order to mix solid APIs and DCIs in NourivanTM/Antiox[®], the producer recommends one of the following wetting agents: water, glycerol, propanediol or sunflower oil [1]. Depending on the active substance added to the base, it may be used to treat, various types of skin lesions. It allows for individualised therapy and more accurate dosing. It has been found that water can be emulsified into the NourivanTM/Antiox[®]. The average value of water

absorbed to 10 g of base using Unguator[®] was 13.18 g. In addition, an attempt was made to determine the amount of water to be added to the base that changes the consistency of the NourivanTM/Antiox[®] base from semi-solid to lotion. The said change was observed after addition of 41.54 g of purified water to 10 g of the base. The vehicle changed consistency to lotion. This makes it possible to use the test base not only as a base for the preparation of semi-solid forms of the drug, but also as a raw material for the formulation of lotions applied by means of e.g. a brush onto dermatological lesions on the skin. The advantage of the base is the cooling effect noticeable after application, associated with the process of water evaporation.

A macroscopic analysis of the prepared formulations confirmed that the base remains homogeneous after mixing in Unguator[®] in a ratio of 0.5:1 with castor oil, Peru balsam and in a ratio of 1:1 with water, 96% ethanol and 86% glycerol, both on the day of preparation and after 4 months from the preparation. When evaluating the application properties of ointment bases, the rheologic parameters should be taken into account. The internal structure of the preparation formed by combining the solid phase and the liquid medium determines its rheological parameters and,

consequently, also the biopharmaceutical parameters of the finished preparation.

The introduction of solvents such as water, castor oil, Peru balsam, ethyl alcohol and glycerol into the NourivanTM/Antiox[®] ointment base results in lower viscosity (Table I). It was proven that an addition of water and glycerol in ratio 1:1 in relation to the base significantly reduces, respectively, two- and four times ($p < 0.01$; $p < 0.001$) its viscosity at the shear rate of 333.3 (1/s). Viscosity of the base with Peru balsam is about two times lower than the viscosity of the base with castor oil, despite the fact that both components were introduced in the same weigh ratios. After introduction of ethyl alcohol, the viscosity of the ointment base significantly decreases ($p < 0.001$) about twenty four times when measured at the shear rate of 333.3 (1/s) and fifteen times when measured at the shear rate of 866.7 (1/s).

The formulations and base themselves were evaluated after 4 months following their preparation. After 4 months of its storage at the temperature of 22°C, NourivanTM/Antiox[®] base is stable, which is visible in slight changes in its viscosity when compared with the values obtained in *ex tempore* tests. Similar observations involve the base with castor oil (Table I).

Table I

Viscosity (mPa·s) of formulations based on NourivanTM/Antiox[®] determined before and after the stability test after 4 months of study

Ointment base + solvent	<i>Ex tempore</i> study		After 4 months of study	
	shear rate 333.3 (1/s)	shear rate 866.7 (1/s)	shear rate 333.3 (1/s)	shear rate 866.7 (1/s)
NA	3404.36 ± 97.11	1389.84 ± 47.01	3310.00 ± 131.05	1440.00 ± 22.16
NA + H ₂ O 1:1	778.72 ± 13.74	423.06 ± 12.32	926.38 ± 47.91*	496.14 ± 24.47*
NA + castor oil 1:0.5	1941.94 ± 65.45	979.40 ± 25.40	2266.56 ± 129.55	1027.24 ± 35.23
NA + Peru balsam 1:0.5	1050.55 ± 115.41	493.78 ± 49.95	513.66 ± 16.06***	272.49 ± 8.66***
NA + ethyl alcohol 1:1	138.56 ± 3.39	95.06 ± 2.74	162.38 ± 3.23*	112.17 ± 3.57*
NA + glycerol 86% 1:1	1270.17 ± 40.42	771.11 ± 26.63	1417.53 ± 20.51*	830.75 ± 8.13*

NA, NourivanTM/Antiox[®]. *($p < 0.05$), **($p < 0.01$), ***($p < 0.001$) as compared to the original values measured *ex tempore*

It was determined that the viscosity significantly ($p < 0.05$) increases when the base is mixed with water, ethyl alcohol and 86% glycerol, respectively, by 19%, 18% and 11.6% at the shear rate of 333.3 (1/s). The base with Peru balsam is the least stable, which is visible in the course of yield curves and viscosity

curves concerning the base mixed with a solvent, immediately after its preparation and after 4 months of its storage. Within the whole range of the shear rates used in the experiment, the viscosity of the base with castor oil and the values of shear stress slightly increased after the storage period (Table II).

Table II

Shear stress (N/m²) of formulations based on NourivanTM/Antiox[®] determined before and after the stability test

Ointment base + solvent	<i>Ex tempore</i> study		After 4 months of study	
	shear rate 333.3 (1/s)	shear rate 866.7 (1/s)	shear rate 333.3 (1/s)	shear rate 866.7 (1/s)
NA	1132.02 ± 32.84	1204.81 ± 40.59	1089.00 ± 22.90	1234.25 ± 10.52
NA + H ₂ O 1:1	259.58 ± 4.59	366.67 ± 10.68	308.84 ± 15.98*	429.98 ± 21.18*
NA + castor oil 1:0.5	647.66 ± 22.30	849.48 ± 22.37	718.44 ± 48.52*	879.72 ± 25.80*
NA + Peru balsam 1:0.5	356.47 ± 35.48	426.00 ± 38.56	172.24 ± 5.32***	234.54 ± 5.97***
NA + ethyl alcohol 1:1	46.35 ± 1.20	82.08 ± 2.34	54.86 ± 1.52*	97.34 ± 2.86*
NA + glycerol 86% 1:1	424.54 ± 14.74	674.44 ± 19.51	472.50 ± 6.79*	720.06 ± 7.07*

NA, NourivanTM/Antiox[®]. *($p < 0.05$), **($p < 0.01$), ***($p < 0.001$) as compared to the original values measured *ex tempore*

When adding the solvents to the ointment base NourivanTM/Antiox[®], the yield stress decreases significantly.

Addition of water lowers the yield stress about seven times ($p < 0.0001$), addition of castor oil – about two

times ($p < 0.05$), of Peru balsam – about four times ($p < 0.01$), of 86% glycerol – about six times ($p < 0.0001$) and an addition of ethyl alcohol- as much as seventy times ($p < 0.0001$) (Table III). The yield limit means the value of shear stress at which the ointment base loses its features of a solid body and becomes liquid. From the application point of view, the lower the yield stress value, the easier it is to smear the preparation on the pathologically changed tissues [19]. The NourivanTM/Antiox[®] ointment base,

with emulsified water, Peru balsam or glycerol 86% shows better extensibility (the values of yield stress, respectively: 109.43; 215.40; 131.57 N/m²) than the base with an addition of castor oil (yield stress of 433.40 N/m²). However, the relatively low value of the yield stress may hinder medicine dosage and cause, among others, a leakage of the preparation from the tube or its flowing down from the spatula. Such situation may involve the base with ethyl alcohol, the yield limit of which is only 4.79 N/m² (Table III).

Table III

Plastic viscosity and yield stress of formulations prepared on the basis of NourivanTM/Antiox[®] determined before and after the stability test

Ointment base + solvent	<i>Ex tempore</i> study			After 4 months of study		
	Plastic viscosity (mPa·s)	Yield stress (N/m ²)	Confidence level of matching (%)	Plastic viscosity (mPa·s)	Yield stress (N/m ²)	Confidence level of matching (%)
NA	45.93 ± 2.58	805.40 ± 11.28	95.46 ± 1.47	45.61 ± 2.43	802.11 ± 10.97	95.36 ± 1.39
NA + H ₂ O 1:1	85.84 ± 4.04	109.43 ± 5.15	98.54 ± 0.24	89.61 ± 2.93*	133.14 ± 9.97**	98.36 ± 1.34
NA + castor oil 1:0.5	76.35 ± 7.32	433.40 ± 9.21	98.11 ± 1.05	79.05 ± 8.57	479.35 ± 8.53	95.61 ± 0.93
NA + Peru balsam 1:0.5	54.75 ± 0.49	215.40 ± 15.34	98.23 ± 0.99	40.36 ± 5.17**	73.20 ± 7.07***	96.51 ± 0.84
NA + ethyl alcohol 1:1	54.06 ± 0.93	4.79 ± 0.13	98.15 ± 1.21	59.20 ± 1.56*	5.74 ± 0.13*	96.85 ± 0.35
NA + 86% glycerol 1:1	238.06 ± 17.34	131.57 ± 9.93	99.15 ± 0.22	247.20 ± 2.86*	157.54 ± 4.95*	98.85 ± 0.21

NA, NourivanTM/Antiox[®]. *($p < 0.05$), **($p < 0.01$), ***($p < 0.001$) as compared to the original values measured *ex tempore*

When testing the yield stress of the bases, after the 4-month long period of storage, it was observed that the NourivanTM/Antiox[®] ointment base showed stable yield stress, which was proven by only slight changes in the critical shear stress, it starts to flow (Table III). A significant increase of the yield stress was ascertained for the base mixed with water - by 21.66% ($p < 0.01$), with ethyl alcohol by 19.83% ($p < 0.05$), and with 86% glycerol by 19.97% ($p < 0.05$), when compared with the original values measured before the stability tests. The ointment base with Peru balsam, after four months of its storage, has got about three times lower value of the yield stress ($p < 0.001$), than immediately after its preparation. The base with ethyl alcohol is also unstable within the yield stress, the value of which changes considerably from 4.79 to 5.74 N/m². During plastic viscosity tests, a significant increase ($p < 0.05$) in plastic viscosity was observed in case of base mixed with water, ethyl alcohol and a significant decrease of the parameter was noted in case of the base mixed with Peru balsam ($p < 0.01$), when compared with the original values measured *ex tempore*.

Conclusions

In the process of drug form formulation, attention should be paid to the responsibility of persons assessing the risk of drug preparation with selected bases used to formulate the drug for the individual patient's needs. Rheological parameters of the bases established by the manufacturer are disturbed by water, castor oil or Peruvian balsam and influence changes in their stability. The direction of changes in stability may also be important. Introduction of the majority

of the solvents used, i.e. water, castor oil, ethyl alcohol, glycerol 86% (w/v) to the base contributes - to a different degree - to an increase of the base viscosity parameters during storage.

An addition of the Peruvian balsam, in turn, reduces the viscosity value and the flow limit of the base over time. The risk assessment of the use of a given base in formulas containing simple ingredients in the everyday recipe practice is also important and necessary. Changes in rheological parameters of the base in the presence of selected formula components will have an impact on the process of releasing therapeutic substances from the formulations, and thus on the therapeutic effect. The observations made can be used to make a decision to combine solvents with other active substances suspended, dissolved, or emulsified. Taking into account the rich composition of the base, further studies are required in order to eliminate potential inconsistencies when using the base as a medium for active substances. The moisturising properties of the base must be emphasised that enables its sole application on the pathologically changed skin, taking into account the active substances contained in the base.

Conflict of interest

The authors declare no conflict of interest.

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