Drug Development and Industrial Pharmacy, 33:1176–1182, 2007

ISSN: 0363-9045 print / 1520-5762 online
DOI: 10.1080/03639040701199274

informa
healthcare

Formulation Compatibility of Myristyl Nicotinate with Drugs Used to Treat Dermatological Conditions Involving an Impaired Skin Barrier

Bassam M. Tashtoush
Department of Pharmaceutical Technology, Faculty of Pharmacy Jordan University of Science and Technology, Irbid-Jordan

Hyuntae Kim, Elaine L. Jacobson, and Myron K. Jacobson
Department of Pharmacology and Toxicology, College of Pharmacy and Arizona Cancer Center, University of Arizona, Tucson, AZ, USA

A number of dermatology conditions including skin photodamage, atopic dermatitis, and rosacea involve skin barrier impairment and first line therapies for these conditions including retinoids and steroids further impair skin barrier function. We have evaluated the compatibility of myristyl nicotinate, an agent that enhances skin barrier function, with drugs used to treat conditions where skin barrier impairment is present including photodamage (retinoic acid), atopic dermatitis (hydrocortisone, triamcinolone acetonide), rosacea (metronidazole), and seborrheic dermatitis (ketoconazole). Myristyl nicotinate was found to be compatible with each of the drugs examined when formulated together and also was shown to be photocompatible with retinoic acid. Our results suggest that the combination of myristyl nicotinate with these drugs is a feasible therapeutic development strategy.

Keywords myristyl nicotinate; retinoic acid; hydrocortisone; triamcinolone; metronidazole; ketoconazole

INTRODUCTION

The skin plays many roles in maintaining homeostasis including providing barrier protection from the environment (Tobin, 2006). The importance of the skin barrier to human health is becoming increasingly apparent as impairment of the barrier function is associated with a number of pathological conditions that include skin photodamage (Bissonauth et al., 1999; Gilchrest, 1996), atopic dermatitis (Hollmann et al., 1988; Park et al., 2006), and rosacea (Tronnier et al., 2004). Additionally, front line therapeutic agents for dermatology conditions including retinoids (Tadaki et al., 1992) and steroids (Fluhr et al., 2003) can cause skin barrier impairment as a side effect of therapy. Myristyl nicotinate, a nicotinic acid derivative developed for optimal delivery of nicotinic acid to skin (Kim et al., 2005). This agent enhances normal epidermal differentiation leading to increased epidermal and stratum corneum thickness with a resulting increase in skin barrier function as determined by significantly reduced rates of transepidermal water loss (Kim et al., 2006b), indicating its potential for treatment of conditions with impaired barrier function and/or to mitigate the side effects of current therapies that impair barrier function. For example, myristyl nicotinate has recently been shown to mitigate skin barrier disruption associated with retinoic acid therapy for skin photodamage (Kim et al., 2006a).

One possible therapeutic development strategy that could capitalize on the skin barrier benefit provided by myristyl nicotinate would be to combine it with agents that are currently used to treat conditions where impaired skin barrier integrity contributes to the condition and/or to combine it with drugs that cause skin barrier impairment to mitigate undesired side effects that result from drug induced barrier impairment. The feasibility of this approach depends upon the compatibility of myristyl nicotinate with a particular drug. Thus, we report here compatibility studies of myristyl nicotinate with drugs currently used for the treatment of dermatology conditions associated with skin barrier impairment including photodamage (retinoic acid), atopic dermatitis (hydrocortisone, triamcinolone acetonide), rosacea (metronidazole), and seborrheic dermatitis (ketoconazole).

MATERIAL AND METHODS

Myristyl nicotinate was provided by Niadyne, Inc. (Tucson, AZ). Tretinoin and trifluoroacetic acid (TFA) were purchased from Sigma Chemical Co. (St. Louis, MO). Acetonitrile and methanol (HPLC grade) were purchased from EMD Chemicals

Address correspondence to Myron K. Jacobson, Department of Pharmacology and Toxicology, College of Pharmacy and Arizona Cancer Center, University of Arizona, Tucson, AZ 85724. E-mail: mjacobson@pharmacy.arizona.edu

Inc. (Gibbstown, NJ). Hydrocortisone, triamcinolone acetonide, ketoconazole, metronidazole, Brij-58, glyceryl monostearate, cetostearyl alcohol, white petrolatum, sorbic acid, butylated hydroxytoluene, simethicone, sorbitol 70% solution, propylene glycol, and polyethylene glycol were purchased from Spectrum Chemical Mfg. Corp. (Gardena, CA). Double distilled deionized water was used.

HPLC Instrumentation and Chromatographic Conditions

An HPLC system consisting of Varian Pro-star solvent delivery system model 230 (Varian Chromatography systems, CA), connected to a UV/Visible Spectroflow 757 absorbance detector (ABI, NJ) and HP 3395 integrator (Hewlett Packard, DE) was used for all separations. The injector was fitted with an injection loop of 50 µl. Chromatographic separation conditions for each compound used in this study are shown in Table 1. All mobile phases were filtered through a 0.45 µm cellulose nitrate membrane filter (Advantec MFS Inc., CA) prior to use. The HPLC assay for each compound was quantified in term of linearity, precision, and accuracy.

Preparation of Dermatological Formulations

Cream formulations of retinoic acid, hydrocortisone, triamcinolone acetonide, ketoconazole, and metronidazole were prepared in the presence and absence of myristyl nicotinate from the following materials as a cream base: water, propylene glycol, sorbitol 70%, sorbic acid, butylated hydroxytoluene, simethicone, white petrolatum, cetostearyl alcohol, Brij-58, glyceryl monostearate, and polyethylene glycol 400. The concentration of myristyl nicotinate and the drugs studied and the ratio of oil phase to aqueous phase for each cream formula are shown in Table 2. The water phase was composed of water, propylene glycol, sorbitol 70%, sorbic acid, butylated hydroxytoluene, and simethicone while the oil phase was composed of white petrolatum, cetostearyl alcohol, Brij-58, glyceryl monostearate, and polyethylene glycol 400. The water phase was mixed and placed in one container at 65–75°C and the oil phase was melted and mixed in another container at 65–75°C. The oil phase was then added to the water phase and mixed until a cream was formed using an IKA mixer model RW 20DZM (IKA-works Inc. NC). Retinoic acid, hydrocortisone, triamcinolone acetonide, and ketoconazole were dissolved in the oil phase while metronidazole was dissolve in the aqueous phase. The cream was cooled to room temperature while stirring.

Extraction of Dermatological Formulations

A sample of each cream formulation was weighed into 50 ml conical centrifuge tube using a Mettler balance model PB-303S (Mettler-Toledo, Switzerland). Myristyl nicotinate was extracted by addition of mobile phase used for HPLC analysis of myristyl nicotinate followed by vortex mixing for 3 min. An aliquot of 1.5 ml was transferred to a microcentrifuge tube

TABLE 1
Chromatographic Conditions for HPLC Analysis of Compounds

Compound	Retinoic Acid	Myristyl Nicotinate	Hydrocortisone	Triamicinolone Acetonide	Ketokonazole	Metronidazole
Column	Nucleosil 5µm C18 100A, 250×4.6 mm (Phenomenex, CA).	Nucleosil 5µm C18 100A, 250×4.6 mm (Phenomenex, CA).	C18 µBondapak,10 µm, 300×3.9 mm (Waters, CA)	C18 µBondapak, 10 µm, 300×3.9 mm (Waters, CA)	C18 µBondapak, 10 µm, 300×3.9 mm (Waters, CA)	C18 µBondapak, 10 µm, 300×3.9 mm (Waters, CA)
Mobile phase	0.01% TFA: Acetonitrile (15:85 v/v)	0.01%TFA: Acetonitrile (2:98 v/v)	Water : Acetonitrile (34:66 v/v)	Water : Acetonitrile (43:57 v/v)	0.01%TFA: Acetonitrile (66:34 v/v)	0.01%TFA: Acetonitrile (85:15 v/v)
Flow rate	1.0 ml/min	2.0 ml/min	1.0 ml/min	1.0 ml/min	1.0 ml/min	0.5 ml/min
Detection wavelength	342 nm	254 nm	254 nm	254 nm	225 nm	315 nm

TABLE 2
Composition of Drug and Myristyl Nicotinate (MN) Formulations

Cream Formulation	MN Concentration % w/w	Drug Concentration % w/w	Oil : Aqueous Phase Ratio
Retinoic acid	0	0.025	27.5:72.5
Retinoic acid and MN	5%	0.025	28.0:72.0
Hydrocortisone	0	1.0	28.5:71.5
Hydrocortisone and MN	10	1.0	30.5:69.5
Triamcinolone acetonide	0	0.1	30.5:69.5
Triamcinolone acetonide and MN	10	0.1	30.5:69.5
Ketoconazole	0	2.0	29.5:70.5
Ketoconazole and MN	10	2.0	32.0:68.0
Metronidazole	0	1.0	28.5:71.5
Metronidazole and MN	10	1.0	32.5:67.5

and subjected to centrifugation at 10,000 rpm for 5 min using an Eppendorf microcentrifuge (Brinkmann Inst Inc., NY). An aliquot of 50 μl of the supernatant was injected directly into the HPLC. The same procedure was followed to extract each of the drugs examined except the extraction solvent was acetonitrile for retinoic acid, and methanol for hydrocortisone, triamcinolone acetonide, and ketoconazole. The extraction solvent for metronidazole was composed of 60% methanol and 40% of the mobile phase used for analysis of metronidazole.

Characterization of Dermatological Formulations

Physical Characterizations

The prepared cream formulations were stored at both room temperature and 40°C for a period of 6 months and at intervals of 2 weeks, the organoleptic characteristics were evaluated for color, texture, phase separation as well as the feel upon application to skin in term of stiffness, grittiness, greasiness, and tackiness. Additionally, a small quantity of each cream was pressed between the thumb and the index finger and the consistency of the cream was noted (whether homogenous or not) and the presence of any particles were examined for by rubbing a small quantity of the cream on the back of the hand.

Chemical Characterizations

For storage stability studies, each of the cream formulations (Table 1) was divided into two parts and placed into tightly closed glass containers and protected from light by covering the storage container with aluminum foil. One container was left at room temperature (22 ± 1°C) and the other container was placed in an oven cabinet at 40 ± 2°C. A sample in triplicate was taken every 2 weeks for up to 6 months and subjected to extraction and analyzed for drug concentration and, if present, myristyl nicotinate concentration.

Photostability Studies

A kilowatt (KW) large area light source solar simulator, model 91293, from Oriel Corporation (Stratford, Connecticut) was used, equipped with 1000W Xenon lamp power supply, model 68920, and a VIS-IR band pass blocking filter plus an atmospheric attenuation filter (output 290–400 nm plus residual 650–800 nm, for SSL). A sample of retinoic acid cream or retinoic acid cream containing myristyl nicotinate was spread uniformly over a cover of 35 mm tissue culture dish and exposed to irradiation with solar simulated light (SSL) at 36.5 cm from the SSL light source. A sample of 0.5 g cream was taken from each cream at selected time intervals, extracted with acetonitrile, and analyzed for retinoic acid as described above.

RESULTS AND DISCUSSION

Physical Characterizations of Drug Formulations

All formulations were examined at the day of preparation and at 2 weeks intervals with regard to gross physical characteristics. All cream formulations remained white, smooth, semisolid, and homogenous with no phase separation at either room temperature or 40°C during the 6 month period. The cream formulations remained free from any detectable particles, they were neither greasy nor tacky, and they spread easily over the skin.

Compatibility of Myristyl Nicotinate with Retinoic Acid

Formulation Compatibility

The chemical compatibility of cream formulations containing retinoic acid alone and in the presence of myristyl nicotinate was investigated by measuring the concentration of retinoic acid and myristyl nicotinate recovered from cream samples stored at both room temperature and 40°C. Retinoic acid cream formulations were stable at room temperature with and without the presence of myristyl nicotinate for the 6 month period of the study (Figure 1A). However, a significant rate of degradation of retinoic acid was observed at 40°C but the presence of myristyl nicotinate decreased the rate of degradation, a difference that was statistically significant ($p < 0.01$).

For example, the formulation containing only retinoic acid was 52.8% degraded after 120 days while the formulation that also contained myristyl nicotinate showed only 15.9% degradation in that time period (Figure 1B). Myristyl nicotinate was shown to be stable when present in retinoic acid cream formulations both at room temperature and 40°C (Figure 1C). These results demonstrate that the presence of myristyl nicotinate is not only compatible with retinoic acid in a cream formulation but also may stabilize the drug in formulations under stressed conditions.

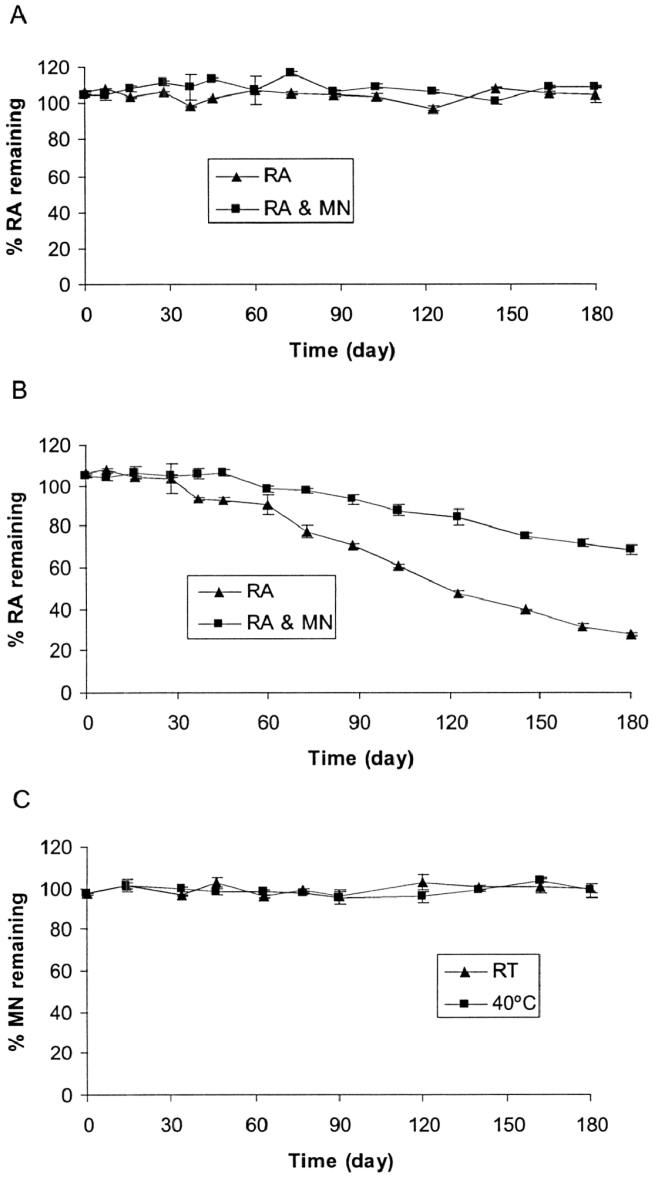

FIGURE 1. Stability of retinoic acid (RA) when formulated alone (triangles) or together with myristyl nicotinate (MN) (squares). Panel A shows formulations stored at room temperature and panel B shows formulations stored at 40°C. Panel C shows MN stability in the formulations containing RA stored at room temperature (triangles) and at 40°C (squares). (N = 3).

Photochemical Compatibility

Retinoic acid is well known to undergo degradation upon exposure to light. Therefore the photocompatibility of myristyl nicotinate with retinoic acid cream was investigated. For that purpose, the photostability of retinoic acid cream formula in the presence and absence of myristyl nicotinate was studied by exposing thin films of creams containing retinoic acid or both retinoic acid and myristyl nicotinate to solar simulated light (SSL) for periods up to 45 min. The rate of degradation was followed by measuring the concentration of retinoic acid as a function of time. The rate of disappearance of retinoic acid in the presence of myristyl nicotinate appeared to be slower than in the formulation containing retinoic acid alone (Figure 2A), although the difference was not statistically significant at the level of p=0.05. The rates of disappearance were examined in more detail by plotting the reciprocal of retinoic acid remaining as a function of time (Figure 2B). This analysis revealed that the degradation of retinoic acid followed second order

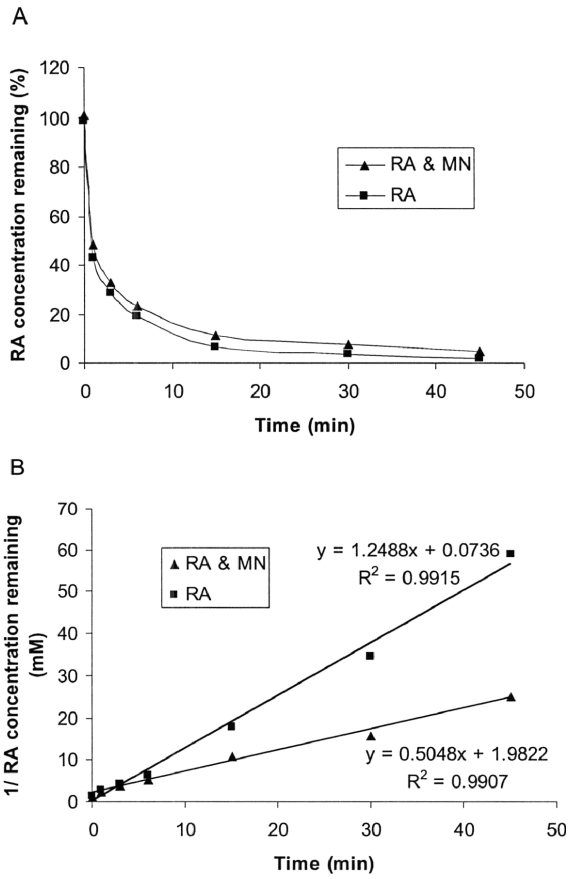

FIGURE 2. Stability of retinoic acid (RA) formulations following exposure to solar simulated light (SSL). Panel A shows RA levels as a function of time for RA when formulated alone (squares) or in the presence of myristyl nicotinate (MN) (triangles). Panel B shows a kinetic analysis via a plot of the reciprocal of RA remaining vs. time for RA alone (squares) or in the presence of MN (triangles). (N = 3).

kinetics and that the rate of photodegradation of retinoic acid was two times higher in the absence of myristyl nicotinate. These results show that myristyl nicotinate is not only photocompatible with retinoic acid but may stabilize the drug following topical application.

Compatibility of Myristyl Nicotinate with Drugs for Atopic Skin Conditions

Compatibility with Hydrocortisone

Hydrocortisone was found to be stable at room temperature in absence and presence of myristyl nicotinate (Figure 3A). At 40°C, the presence of myristyl nicotinate showed a statistically significant ($p < 0.05$) stabilizing effect as a cream containing hydrocortisone alone lost 18.2% after 6 months of storage while the formulation containing only myristyl nicotinate lost 4.5% (Figure 3B). Myristyl nicotinate was found to be stable in formulations containing hydrocortisone at both room temperature and 40°C (Figure 3C). These results indicate that myristyl nicotinate is compatible with hydocortisone in cream formulations and that its presence may stabilize the drug under stressed conditions.

Compatibility with Triamcinolone Acetonide

Triamcinolone acetonide was stable in absence and presence of myristyl nicotinate at both room temperature and 40°C during the study period (Figure 4A, 4B). There were no

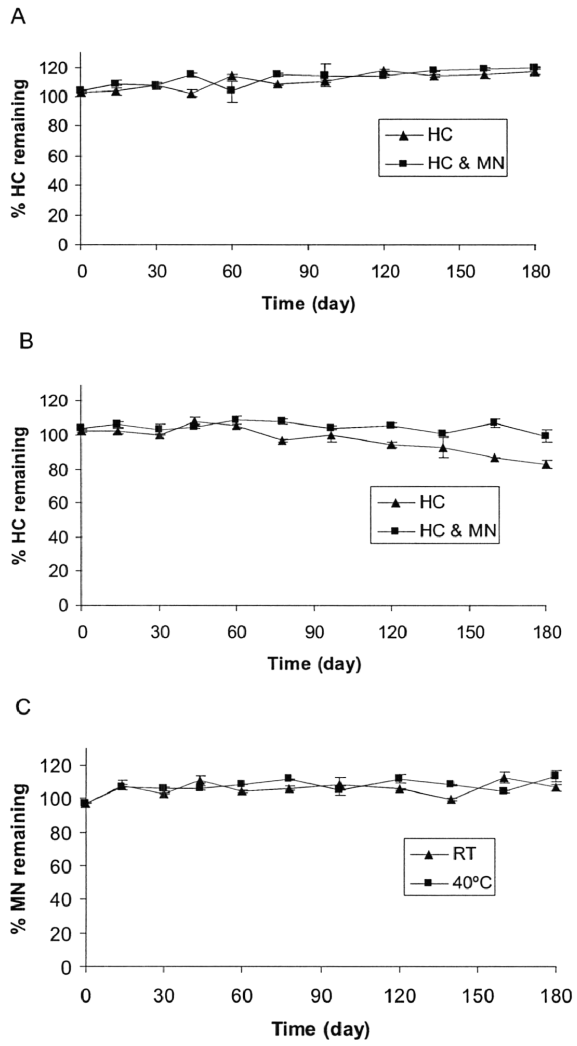

FIGURE 3. Stability of hydrocortisone (HC) when formulated alone (triangles) or together with myristyl nicotinate (MN) (squares). Panel A shows formulations stored at room temperature and panel B shows formulations stored at 40°C. Panel C shows MN stability in the formulations containing HC stored at room temperature (triangles) and at 40°C (squares). ($N = 3$).

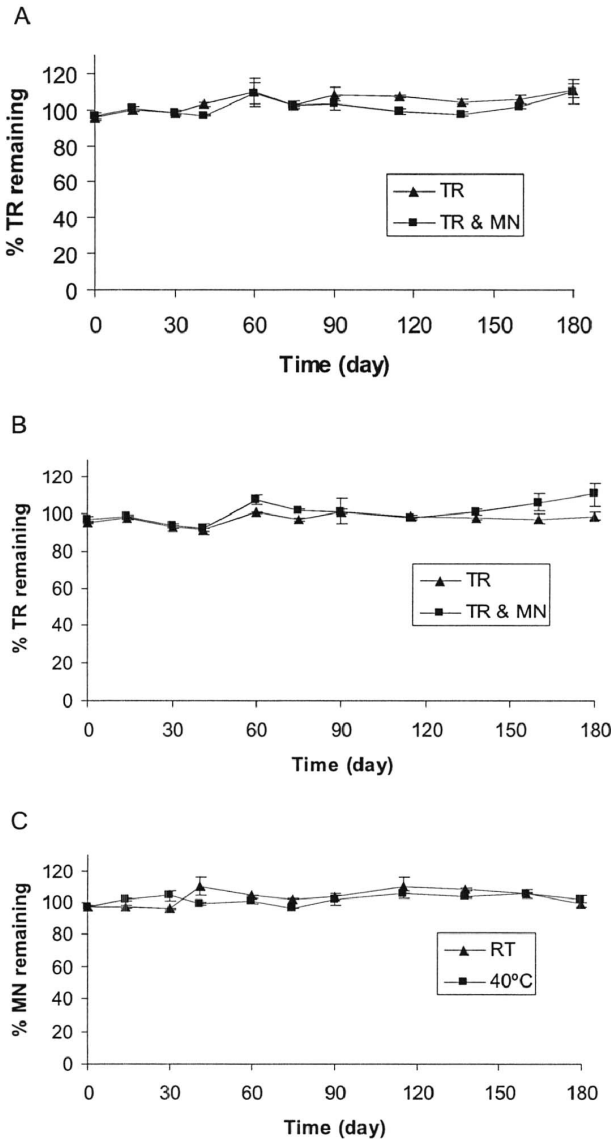

FIGURE 4. Stability of triamcinolone acetonide (TR) when formulated alone (triangles) or together with myristyl nicotinate (MN) (squares). Panel A shows formulations stored at room temperature and panel B shows formulations stored at 40°C. Panel C shows MN stability in the formulations containing TR stored at room temperature (triangles) and at 40°C (squares). ($N = 3$).

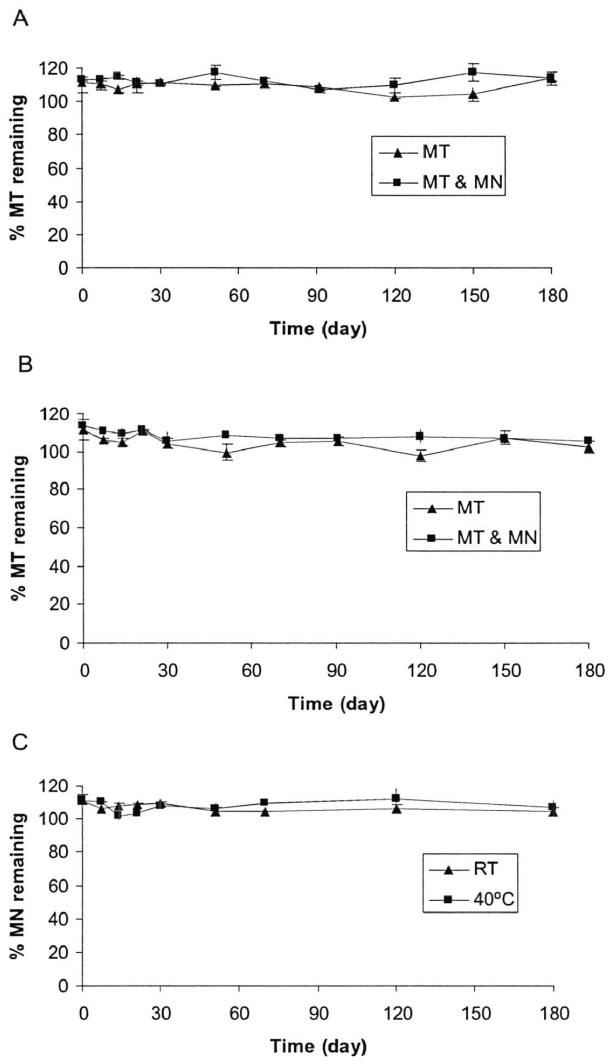

FIGURE 5. Stability of metronidazole (MT) when formulated alone (triangles) or together with myristyl nicotinate (MN) (squares). Panel A shows formulations stored at room temperature and panel B shows formulations stored at 40°C. Panel C shows MN stability in the formulations containing MT stored at room temperature (triangles) and at 40°C (squares). (N = 3).

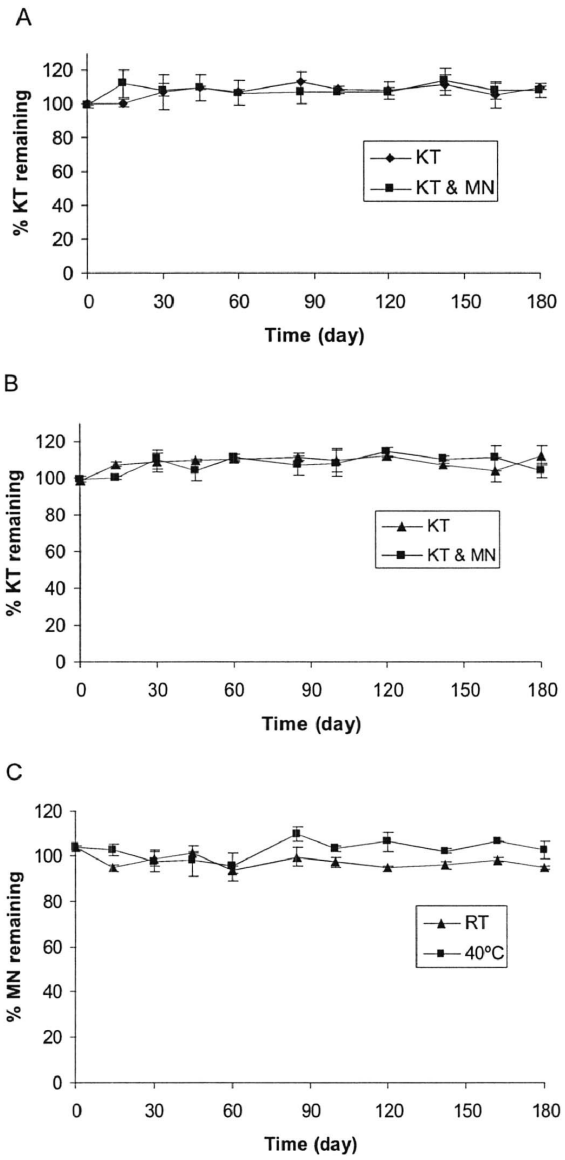

FIGURE 6. Stability of ketoconazole (KT) when formulated alone triangles) or together with myristyl nicotinate (MN) (squares). Panel A shows formulations stored at room temperature and panel B shows formulations stored at 40°C. Panel C shows MN stability in the formulations containing KT stored at room temperature (triangles) and at 40°C (squares). (N = 3).

statistically significant differences that approached a p value of 0.05. Furthermore, myristyl nicotinate was stable in the triamcinolone acetonide cream formulation (Figure 3C). These results indicate that triamcinolone acetonide and myristyl nicotinate are compatible in dermatological formulations.

Compatibility of Myristyl Nicotinate with Drugs for Rosacea

Compatibility with Metronidazole

Metronidazole showed no detectable degradation at either room temperature or 40°C in the absence or presence of myristyl nicotinate (Figure 5A, 5B) and myristyl nicotinate was found to be stable in metronidazole cream formulation at the two test conditions (Figure 5C), indicating formulation compatibility of myristyl nicotinate with metronidazole.

Compatibility with Ketoconazole

Ketoconazole showed no detectable degradation at either room temperature or 40°C in the absence or presence of myristyl nicotinate (Figure 6A, 6B) and myristyl nicotinate was found to be stable in ketoconazole cream formulation at the two test conditions (Figure 6C), indicating formulation compatibility of myristyl nicotinate with ketoconazole.

CONCLUSIONS

Myristyl nicotinate is compatible in dermatological formulations with retinoic acid, hydrocortisone, triamcinolone acetonide, metronidazole, and ketoconazole. The compatibility of myristyl nicotinate with drugs used to treat conditions where skin barrier impairment contributes to the condition demonstrates that a drug development strategy combining myristyl nicotinate with existing drugs is a feasible therapeutic development strategy for improving the efficacy of treatment of skin photodamage, atopic dermatitis, rosacea, and seborrheic dermatitis. Combining myristyl nicotinate with retinoic acid is also a feasible strategy to reduce side effects of treatment of skin photodamage and acne. Combining myristyl nicotinate with hydrocortisone or triamcinolone acetonide to reduce drug related barrier impairment also appears to be a feasible development strategy.

ACKNOWLEDGMENTS

These studies were funded in part by Niadyne, Inc., and NIH grant R44 CA-90085. MKJ and ELJ are principals in Niadyne, Inc., whose sponsored research is managed in accordance with the University of Arizona conflict-of-interest policies.

REFERENCES

Dirschka, T., Tronnier, H., & Folster-Holst, R. (2004). Epithelial barrier function and atopic diathesis in rosacea and perioral dermatitis. *Br. J. Dermatol., 150*(6), 1136–1141.

Gilchrest, B. A. (1996). A review of skin ageing and its medical therapy. *Br. J. Dermatol. 135*(6), 867–875.

Jacobson, E. L., Kim, H., Coyle, W. R., Kim, M., Coyle, D. L., Rizer, R., Jaconson, M. K. (In Press). Effect of myristyl nicotinate on retinoic acid therapy for facial photodamage. *Experimental Dermatology*.

Jacobson, E. L., Kim, H., Kim, M., Williams, J. D., Coyle, D. L., Coyle, W. R., Stratton, M. S., Grove, G., Rizer, R., & Jaconson, M. K. (2006). A topical lipophillic niacin derivative increases NAD, epidermal differentiation and skin barrier function in photodamaged skin. *Experimental Dermatology, 16*, 490–499.

Jacobson, E. L., Kim, H., Kim, M., Wondrak, G. T., & Jacobson, M. K. (2005). Developing Topical Prodrugs for Skin Cancer Prevention. In, D. S. Alberts, L. M. Hess, *Fundamentals of Cancer Prevention* (pp. 139–160). Berlin Heidelberg: Springer-Verlag.

Kao, J. S., Fluhr, J. W., Man, M. Q., Fowler, A. J., Hachem, J. P., Crumrine, D., Ahn, S. K., Brown, B. E., Elias P. M., & Feingold, K. R. (2003). Short-term glucocorticoid treatment compromises both permeability barrier homeostasis and stratum corneum integrity: Inhibition of epidermal lipid synthesis accounts for functional abnormalities. *J. Invest. Dermatol. 120*(3), 456–464.

Kim, D. W., Park, J. Y., Na, G. Y., Lee, S. J., & Lee, W. J. (2006). Correlation of clinical features and skin barrier function in adolescent and adult patients with atopic dermatitis. *Int. J. Dermatol. 45*(6), 698–701.

Melnik, B., Hollmann, J., & Plewig, G. (1988). Decreased stratum corneum ceramides in atopic individuals—A pathobiochemical factor in xerosis? *Br. J. Dermatol. 119*(4), 547–549.

Robert, M., Bissonauth, V., Ross, G., & Rouabhia, M. (1999). Harmful effects of UVA on the structure and barrier function of engineered human cutaneous tissues. *Int. J. Radiat. Biol., 75*(3), 317–326.

Tagami, H., Tadaki, T., Obata, M., & Koyama, J. (1992). Functional assessment of the stratum corneum under the influence of oral aromatic retinoid (etretinate) in guinea-pigs and humans. Comparison with topical retinoic acid treatment. *Br. J. Dermatol., 127*(5), 470-475.

Tobin, D. J. (2006). Biochemistry of human skin—Our brain on the outside. *Chem. Soc. Rev. 35*(1), 52–67.