Journal of Oil Palm Research Vol. 24 April 2012 p. 1287-1295

ZINC GLYCEROLATE: POTENTIAL ACTIVE FOR TOPICAL APPLICATION

CHEONG MEI YEE*; HAZIMAH ABU HASSAN*; ZAFARIZAL ALDRIN AZIZUL HASSAN* and ROSNAH ISMAIL*

ABSTRACT

Crystalline metal-based glycerol complexes, such zinc (Zn), cobalt (Co), iron (Fe), nickel (Ni) and manganese (Mn) complexes, are formed by heating certain metal oxides, hydroxides or salts with glycerol at temperatures above 110°C. In this study, zinc glycerolate were synthesised using zinc oxide and excess glycerol at 240°C. The characterisations done were Fourier transform infrared spectroscopy (FTIR), thermogravimetric analysis and differential thermal analysis (TGA-DTA), scanning electron microscopy (SEM), X-ray diffraction, in vitro *dermal irritection assay and determination of sun protection factor (SPF). The hydrolysis property of zinc glycerolate in aqueous condition was also investigated. The hydrolysis of zinc ions followed a rather simple path unlike in the case of most polyvalent cations. The reaction of Zn^{2+} with OH^- ions resulted first in the formation of $Zn(OH)^+$ species followed by precipitation of zinc hydroxide which easily redissolved in an excess of base to form a soluble complex. Quantitative TGA-DTA analysis was used to determine the amount of glycerol required in aqueous solution that could presumably minimise the hydrolysis of zinc glycerolate. Aqueous solutions that were subjected to high heat needed up to 60% of glycerol in order to preserve the zinc glycerolate. Therefore based on this study, zinc glycerolate is more suited to be incorporated in dry or anhydrous formulations such as powders and ointments. It has SPF of 1.07 ± 0.004 and ultraviolet A (UVA) ratio of 0.29, which is a moderate (one star) Boots star rating for UVA protection. Zinc glycerolate is classified as non-irritant, and therefore has good potential for use in the pharmaceutical and personal care product industries.*

Keywords: zinc glycerolate, metal-glycerol complex.

Date received: 10 November 2010; **Sent for revision:** 16 November 2010; **Received in final form:** 17 March 2011; **Accepted:** 2 February 2012.

INTRODUCTION

Glycerol is an important oleochemical commodity with its production greatly stimulated by the recent popularity of biodiesel. Glycerol forms complexes with many metal ions. As first disclosed by Radoslovich *et al.* in 1970, heating certain metal oxides, hydroxides or salts with glycerol at temperatures above 110°C yielded crystalline metal complexes. The metals include cobalt, iron, manganese and zinc, and the complexes formed differ from those obtained by reaction in aqueous solution at room temperature in that they are mostly insoluble in water and organic solvents (Radoslovich *et al.,* 1970; Brylants and Phillippe, 1980; Hambley and Snow, 1983; Mendolovici *et al.*, 1986; Nargy *et al.*, 1986; Taylor and Brock, 1989; Whitehouse *et al.,* 1990). It has been reported that glycerol through a copper-mediated reaction (copper-glycerol complex) can be an intermediate product for specialty chemicals (Hazimah *et al.*, 2001). Of all the metal-based glycerol complexes, zinc glycerolate has many uses, including for the treatment and prevention of ammoniacal dermatitis, pruritis, psoriasis, tinea pedis and industrial dermatitis (Taylor and Brock., 1985).

Zinc glycerolate, zinc monoglycerolate ($C_3H_6O_3Zn$) or, more correctly,

* Malaysian Palm Oil Board,
P. O. Box 10620, 50720 Kuala Lumpur, Malaysia.
E-mail: cheongmy@mpob.gov.my

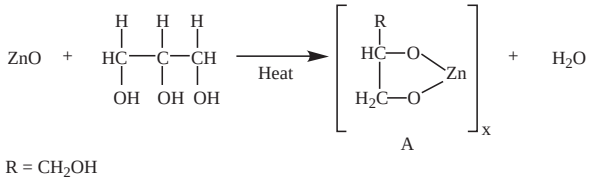

Figure 1. General glyceration reaction between glycerol and zinc oxide.

zinc [1,2,3-propanetriolato(2-)-O_1,O_2] is formed by heating zinc oxide with glycerol at 120°C-300°C as shown in *Figure 1*. The empirical formula for the zinc glycerolate complex is $(C_3H_6O_3Zn)_x$ (Whitehouse *et al*., 1990). Zinc glycerolate can be prepared by the simple conventional method and by the microwave radiation method (Taylor, 1990). Basically, the preparation of zinc glycerolate entails mixing zinc oxide with excess glycerol at about 260°C, maintaining the temperature with constant stirring until the reaction is completed, followed by cooling, washing with water, filtering and drying to isolate the solids. After removing excess glycerol and moisture, the product has a talc-like property with good skin covering power and adhesion. As expected, the reaction will proceed more slowly at lower temperatures.

In 2006, Taylor and co-inventors patented a method for producing microfine zinc glycerolate complexes. Generally, the method involves reacting a salt form of zinc, preferably zinc acetate with an excess of glycerol under controlled temperature, pressure and agitation for a time sufficient to effectuate reaction between the dissolved zinc and the glycerol. Generally, temperature, pressure, overall molar ratio of glycerol to zinc, rate of agitation and sufficient time to effectuate reaction between glycerol and dissolved zinc are the driving factors in controlling the size of the zinc glycerolate particulates. The patent also revealed that the molar ratio of glycerol to zinc in the starting material ranges from 2:1 to about 10:1, preferably from about 3:1 to 4:1 (Taylor *et al*., 2006). It is also reported in literature that the method of formation can be improved by subjecting a suspension of zinc oxide in an excess of glycerol to microwave radiation in a microwave reactor. Improvement in the quality of the crystalline zinc glycerolate complex formed may be demonstrated by electron micrographs. Microwave action induces significant vibration in particular molecular bonds which accelerates and enhances the reaction (Taylor, 1990).

The morphology of the zinc oxide source has a pronounced effect on the appearance of the zinc glycerolate product. Remias *et al.* in 2009 synthesised 2 to 4 µm long zinc glycerolate microstacks in which 6-12 hexagonal prisms were aligned face to face through treating nanorod-shaped zinc oxide with glycerol at 100°C under reflux. The nanorod-shaped zinc oxide was first obtained by the thermal decomposition of zinc oxalate nanorods. In contrast, without any structure-directing effects the product zinc glycerolate was obtained as a random heap of hexagonal prisms with an average diameter and thickness of ca. 2.5 µm and ca. 350 nm, respectively.

Zinc glycerolate obtained through the conventional direct heating of zinc oxide and glycerol has a platy morphology with generally hexagonal crystals of relatively low thickness and high lubricity. It is insoluble in water and common organic solvents, exhibits a hydrophobic characteristic, and retards the growth of fungi and some bacteria (Taylor and Brock, 1985). X-ray crystallographic analysis of the zinc glycerolate revealed that the crystals are monoclinic (Hambley and Snow, 1983).

Zinc glycerolate is also useful as a modifier of plastic or rubber materials because it can improve their physical properties by increasing tensile strength and reducing ultraviolet light degradation of these organic materials (Taylor, 1988). However, the notable uses of zinc glycerolate are in cosmetics, pharmaceutical and therapeutic applications. This includes the treatment of local skin disorders and systemic inflammatory disease like arthritis. The therapeutic benefits of zinc glycerolate come from Zn^{2+} of the zinc and glycerol complex when rubbed on the skin or used *in vivo*. When ^{65}Zn marker used in zinc glycerolate was applied to the skin of rats, the marked Zn appeared in the excreta, indicating significant dermal absorption of zinc from this complex (Fairlie *et al.*, 1992). Several studies have been reported by Taylor and Brock (1989) which show that percutaneous, subcutaneous, intramuscular and oral administrations of zinc glycerolate are useful in the treatment of diabetes. It is an anti-microbial or anti-bacterial agent and an anti-inflammatory agent in the treatment of arthritis and psoriasis. It can also be used for the treatment of zinc deficiency, and as a gastroprotective agent in the treatment of gastric ulcers. There is also a study on the use of zinc glycerolate in the treatment of oral herpetic sores (Apisariyakulm *et al.*, 1990). There is potential in the *in vivo* use of zinc glycerolate as an effective hypoglycemic agent in prevention and treatment of diabetes as this complex has low toxicity and high insulin-like activity (Coulston and Dansona, 1980; May and Contoreggi, 1982; Shisheva *et al.*, 1992).

This article reports the straightforward synthesis and characterisation of zinc glycerolate. The stability of zinc glycerolate in aqueous and glycerol solutions by quantification of thermal gravimetric analysis is discussed. *In vitro* dermal irritation assay was used to assess its irritation potential on human skin. The sun protection factor (SPF) of zinc glycerolate was determined via an ultraviolet transmittance analyser.

MATERIALS AND METHODS

Materials

Zinkoxid Reinst (ZnO) was obtained from Merck while analytical grade glycerol was obtained from Fisher Scientific. Acetone was obtained from J T Baker for use in the purification of the samples. All other reagents were of analytical grade and were used as received.

Preparation Methods

Zinc oxide and glycerol at a molar ratio of 1:3 were heated with frequent agitation using an Ethos MR microwave reactor (Milestone, Germany). The initial duration of heating and temperature were set at 10 min and 40ºC, respectively. The optimum temperature was set at 250ºC, and the retention time at this temperature was varied from 20 min to 1 hr. The venting time was set at 10 min. The reaction mixture was left to cool, and later purified using acetone. The slurry was filtered using a Buchner funnel lined with filter paper No. 1 from Whatman. The sample was dried in a conventional oven at 90ºC for 20 min.

Characterisation

The crystalline structure of the dried solids was evaluated by X-ray powder diffraction with a Rigaku TTRAX III high power simultaneous X-ray diffraction system with differential scanning calorimetry (DSC) using Cu*K* α (λ = 1.54056 Å) as incident radiation. The morphology of the zinc glycerolate particles were scanned by a Philips XL30 scanning electron microscope. The particle size of dispersed zinc glycerolate was obtained by laser diffraction size analysis using a Malvern Mastersizer Hydro 2000S particle size analyser. FTIR spectra were recorded by Nicolet, Magna-IR 550 spectrophotometer, Series II, USA. FTIR spectra were obtained by grinding the samples with potassium bromide (KBr) before compressing them into pellets. In investigating the optimum quantity of glycerol to be included in the aqueous solution to minimise hydrolysis, TGA-DTA of the samples was carried out and recorded on a SDT 2960 simultaneous DSC-TGA thermal analyser. The samples were heated to 1000ºC at a heating rate of 10°C per minute under nitrogen gas, and at a flow rate of 50-60 ml per minute. The samples were cooled using air. The TGA-DTA residues were kept for FTIR analysis.

In vitro ***dermal irritation assay of zinc glycerolate.*** This compound may be used as an active in cosmetic formulations, and hence must cause minimal skin irritation. The principle of the testing is as follows: dermal irritancy induced by any substance or ingredient is related to its ability to denature and disrupt corneal proteins or alter the structure of keratin, collagen and other dermal proteins. The dermal irritection assay (Anon., 1996), an alternative method to animal irritancy tests or Draize test, correlates well with *in vivo* irritancy tests (Sina *et al.*, 1995). Dermal irritection assay requires two components: a membrane substrate modified by covalently cross-linking a mixture of keratin, collagen and an indicator dye, and a reagent solution consisting of an organised globulin/protein matrix. The changes in protein structure induced by the test material are readily quantified by the changes in turbidity at 405 nm (OD_{405}). Application of an irritant chemical to the membrane disc disrupts the ordered structure of keratin and collagen, and releases the bound indicator dye. Comparison of its optical density to those produced by standard chemical irritants permits the calculation of an irritancy score that has been shown to be directly related to the dermal irritancy of the test material. The dermal irritancy potential is expressed in a Human Irritancy Equivalent (HIE) score. The predicted *in vivo* classifications based on these scoring systems are shown in *Table 1*.

Samples were weighed at five different concentrations: 25, 50, 75, 100 and 125 mg. These were placed into the membrane discs. The reagent and blanking buffer (1250 µl) were added to a 24-well assay plate. The membrane discs that contained various concentrations of zinc glycerolate sample were inserted into the corresponding blank and test sample wells of the plate. The assay plate was then incubated at 25°C for 24 hr. Then the membrane discs were removed from the assay plate, and subsequently 250 µl of the reagent and blanking buffer were transferred into a 96-well reading plate. This plate was inserted into a MRX microplate reader (Dynex Technologies, Inc. Chantilly, VA).

In vitro ***SPF determination of zinc glycerolate.*** A Labsphere ultraviolet transmittance analyser UV-1000S was used to evaluate the SPF value of zinc glycerolate. The 3M Transpore™ tape was used as the substrate for the *in vitro* SPF measurement because of its transparency to ultraviolet light,

TABLE 1. CLASSIFICATION OF HUMAN IRRITANCY EQUIVALENT SCORE IN RELATION TO *in vivo* IRRITANCY CLASSIFICATION

Human irritancy equivalent	Predicted dermal irritancy classification
0 – 0.90	Non-irritant
0.90 – 1.20	Non-irritant/Irritant
1.20 – 5.00	Irritant

simulating the porosity and texture of human skin. The tape had uneven topography that enabled the distribution of the test material as a sunscreen in a way similar to human skin. The 3M Transpore™ tape was placed in a single layer on clean 2-mm thick quartz slides. An area of at least two square inches (12.5 cm^2) was used to enable measurements over at least five non-overlapping spots. Samples of 2 mg cm^{-2} were distributed evenly on the entire surface of the sample plate. The samples were put aside to dry, and the emulsion allowed to break for at least 20 min before measurements were recorded. The testing of samples was performed by running a baseline on the reference media before the prepared samples were analysed.

SPF by definition is determined *in vivo* as an increase in exposure time is required to induce erythema. However, the *in vitro* technique involved measuring the spectral transmittance at UV wavelengths from 280 to 400 nm. The *in vitro* SPF is calculated as follows:

$$SPF = \frac{\int_{280nm}^{400nm} E_\lambda . S_\lambda d\lambda}{\int_{280nm}^{400nm} E_\lambda . S_\lambda . T_\lambda . d\lambda}$$

where,

E_λ = CIE erythermal spectral effectiveness.

S_λ = solar spectral irradiance.

T_λ = spectral transmittance on the sample (as measured on UV-1000S).

The equation shows that the higher the amount of transmittance, the lower the SPF value. The transmittance spectrum of a sunscreen in either region is averaged in order to produce one value, which describes the amount of UV-A or UV-B blocking. The average transmittance in each region is given by:

$$T\ (UVA)_{av} = \frac{\sum_{315nm}^{400nm} T_\lambda \Delta_\lambda}{\sum_{315nm}^{400nm} \Delta_\lambda}$$

and

$$T\ (UVB)_{av} = \frac{\sum_{280nm}^{315nm} T_\lambda \Delta_\lambda}{\sum_{280nm}^{315nm} \Delta_\lambda}$$

where, Δ_λ = measured wavelength interval.

Consequently, the percent blocking for UVA and UVB, respectively, is calculated as;

100% - $T\ (UVA)_{av}$ or 100% - $T\ (UVB)_{av}$

where, $T(UVA)_{av}$ or $T(UVB)_{av}$ is expressed as a percentage.

In addition to its ability to determine SPF of a sunscreen, the *in vitro* technique can also measure the UVA protection of the sunscreen. Boots the Chemist, the largest producer of sunscreens in the United Kingdom, has developed a label system that uses a four star rating based on spectrophotometric analysis. The Boots star rating system is a proprietary *in vitro* method used to describe the ratio of UVA to UVB protection offered by sunscreen creams and sprays as classified in *Table 2*.

TABLE 2. BOOTS STAR RATING AND CLASSIFICATION FOR ULTRAVIOLET A (UVA) PROTECTION DETERMINED FROM MEASURED UVA RATIOS

UVA ratio	Star category	Category descriptor
0.0 to < 0.2	-	Too low for UVA claim
0.2 to < 0.4	*	Moderate
0.4 to < 0.6	**	Good
0.6 to < 0.8	***	Superior
≥ 0.8	****	Maximum

The spectral transmittance values, T_λ, were converted to spectral absorbance values, $A_\lambda = -\log(T_\lambda)$. A term called the UVA ratio was calculated, which is the ratio of the total absorption in UVA to that in UVB.

$$\frac{aUVA}{aUVB} = \frac{\int_{280nm}^{400nm} A_\lambda d\lambda}{\int_{290nm}^{320nm} A_\lambda . d\lambda}$$

RESULTS AND DISCUSSION

The scanning electron microscopy of zinc glycerolate and zinc oxide are given in *Figures 2a* and *2b*, respectively. The large, plate-like and hexagonal shape of the zinc glycerolate complex was well defined; which distinguished it easily from the more regular fine particulates of zinc oxide. Zinc glycerolate which was formed had a substantially greater development in two dimensions in one plane than in a third dimension normal to the other plane. The lustrous, large sized crystallite particles endowed zinc glycerolate with a high lubricity which is an advantageous attribute for topical dermal application (Taylor and Brock, 1989).

The average particle size by volume weighted mean D [4,3] of zinc glycerolate was 23.015 µm as shown in *Figure 3* while for zinc oxide it was 12.543 µm. *Figure 4* shows the Fourier transform infrared spectrocopy (FTIR) spectrum of a purified zinc glycerolate sample. The appearance of strong and broad absorption bands in the region of 3600 to 3000 cm^{-1} and a medium-sharp absorption band at 1650-1600 cm^{-1} usually comes from the stretching and bending vibration modes, respectively, of the free hydroxyl (OH) group. This indicated the minute amount of water present in the complex samples or unreacted free glycerol that was left in the samples. The three strong and sharp absorption bands at 2950-2850 cm^{-1} were due to C-H stretching vibration modes. The sharp absorption bands at 1120-990 cm^{-1} are assigned to C-O bending modes of alcohol present in the complex samples (Lide, 1994; Pavia *et al.*, 1996).

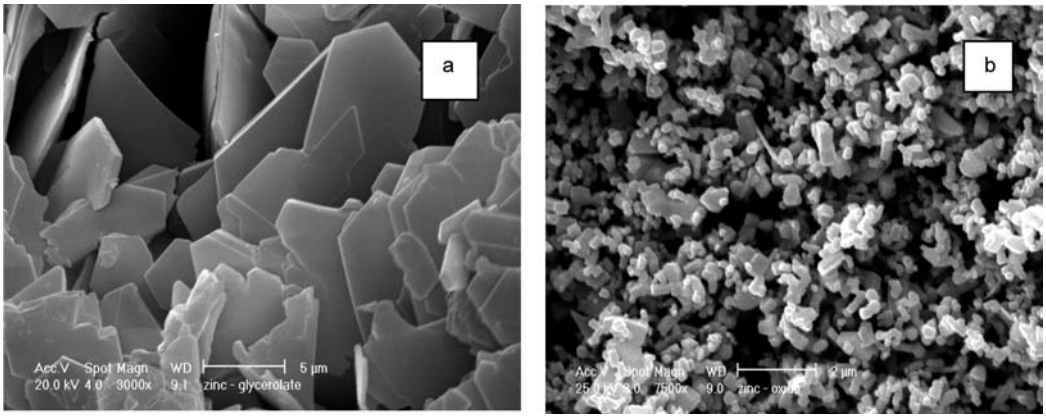

Figure 2. Scanning electron microscopy of (a) zinc glycerolate magnified to 3000X and (b) zinc oxide magnified to 7500X.

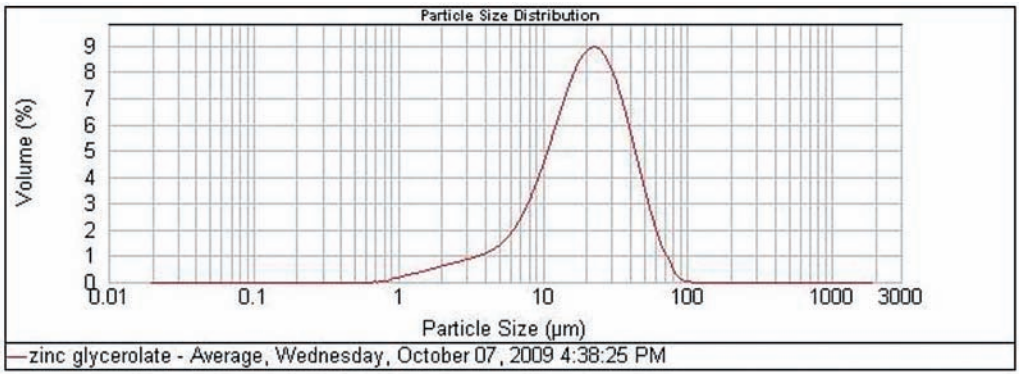

Figure 3. Size distribution of zinc glycerolate particles obtained.

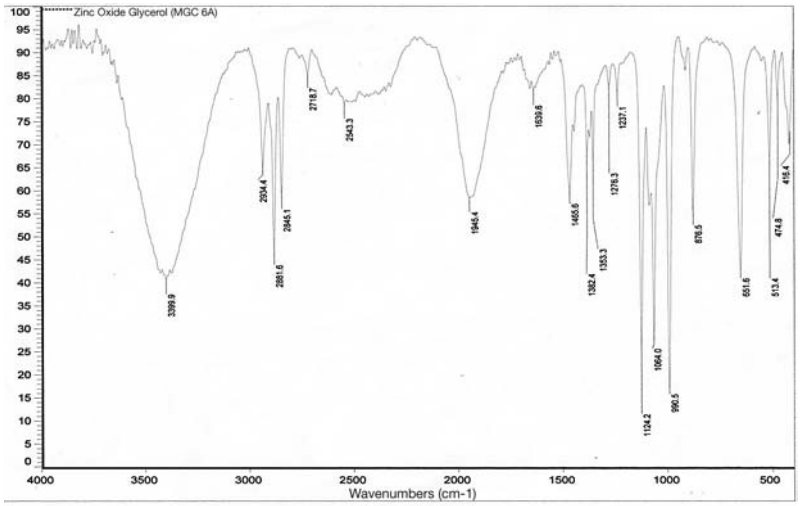

Figure 4. The Fourier transform infrared spectoscopy (FTIR) spectrum of zinc glycerolate.

Absorptions in the fingerprint region from 1300 to 850 cm^{-1} included contributions from complex interacting vibrations, giving rise generally to a unique fingerprint for each compound. The fingerprint region was too intricate to interpret and was almost similar to that of glycerol. The absorption bands below 550 cm^{-1} were assigned to metal-ligand complexes which in this case indicate the zinc glycerolate complex.

The zinc glycerolate TGA-DTA thermogram indicated many successive mass losses. The presence of absorbed water molecules and solvents was shown by the mass loss below 150ºC, while the weight loss from 150ºC to 1000ºC can be attributed to the ligand dissociation. Free glycerol was accounted for by the mass loss below 290ºC, while mass loss between 290ºC and 1000ºC can be attributed to the glycerolate ($C_3H_6O_2^-$) ions.

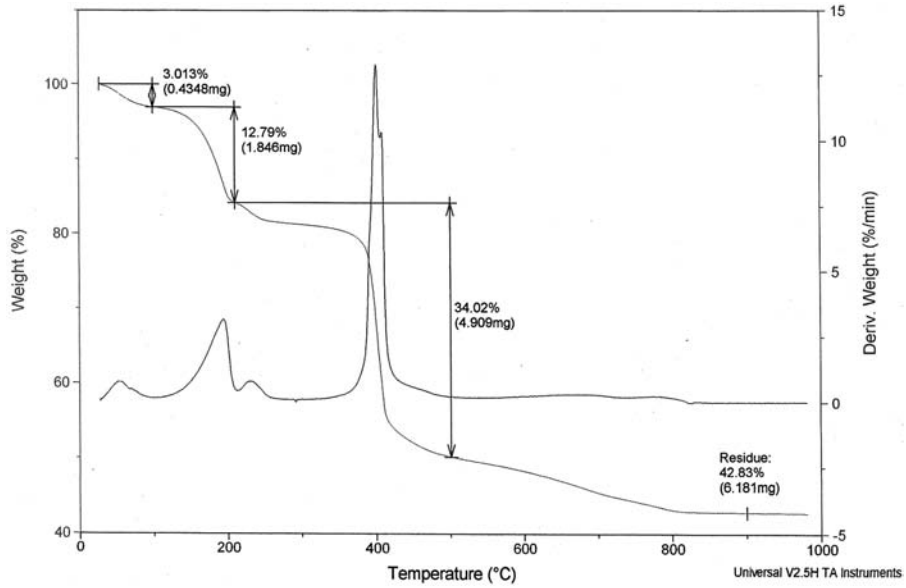

Figure 5. The thermogravimetric analysis and differential thermal analysis (TGA-DTA) thermogram profile of a zinc glycerolate sample.

Zinc glycerolate had final residues of zinc oxide as its calcination final product. The purity of zinc glycerolate can be quantified by correlating each peak and weight loss curve to the pyrolised material in the thermogram. Thus, in *Figure 5*, the percentage of the zinc glycerolate complex in the sample was 92.96% while the percentage of residue as zinc oxide was 7.04%. *Figure 6* shows the X-ray diffraction profile of a zinc glycerolate sample.

Figure 7a shows an interesting change had occurred in the zinc glycerolate complex sample that was stored in aqueous solution for three months. The TGA-DTA thermogram of the hydrolysed zinc glycerolate complex showed a profile similar to that of zinc oxide in *Figure 7b*. The hydrolysis phenomenon of the zinc glycerolate complex in aqueous solution to zinc oxide was proven by the X-ray diffraction analysis in *Figures 8a* and *8b* which showed distinctly similar profiles.

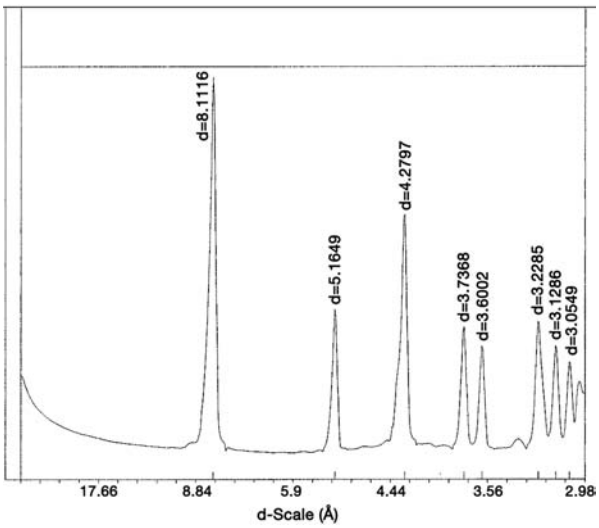

Figure 6. The X-ray diffraction profile of a zinc glycerolate sample.

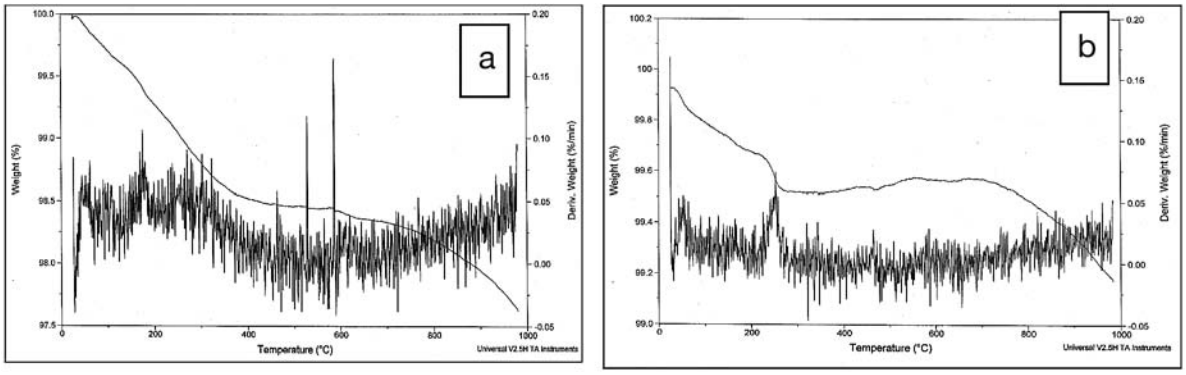

Figure 7. The thermogravimetric analysis and differential thermal analysis (TGA-DTA) thermogram profiles of (a) zinc glycerolate that had been stored for three months in aqueous solution, and (b) zinc oxide.

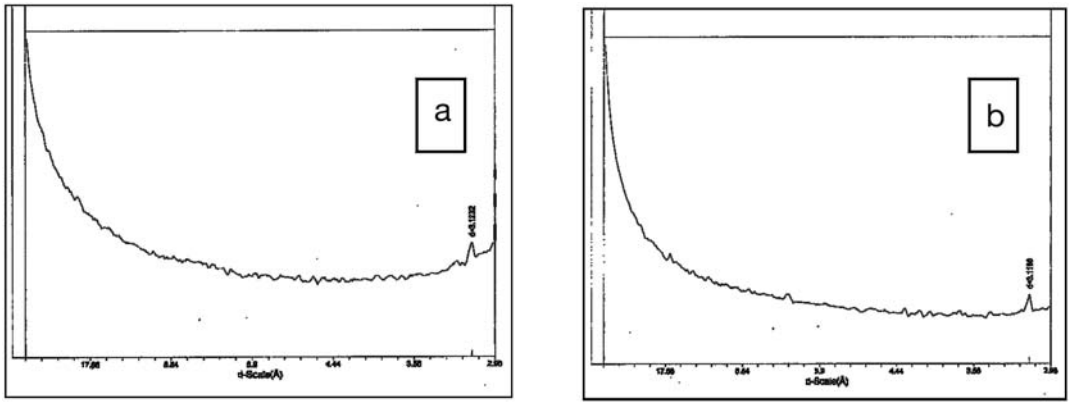

Figure 8. The X-ray diffraction profiles of (a) zinc glycerolate that had been stored for three months in aqueous solution, and (b) zinc oxide.

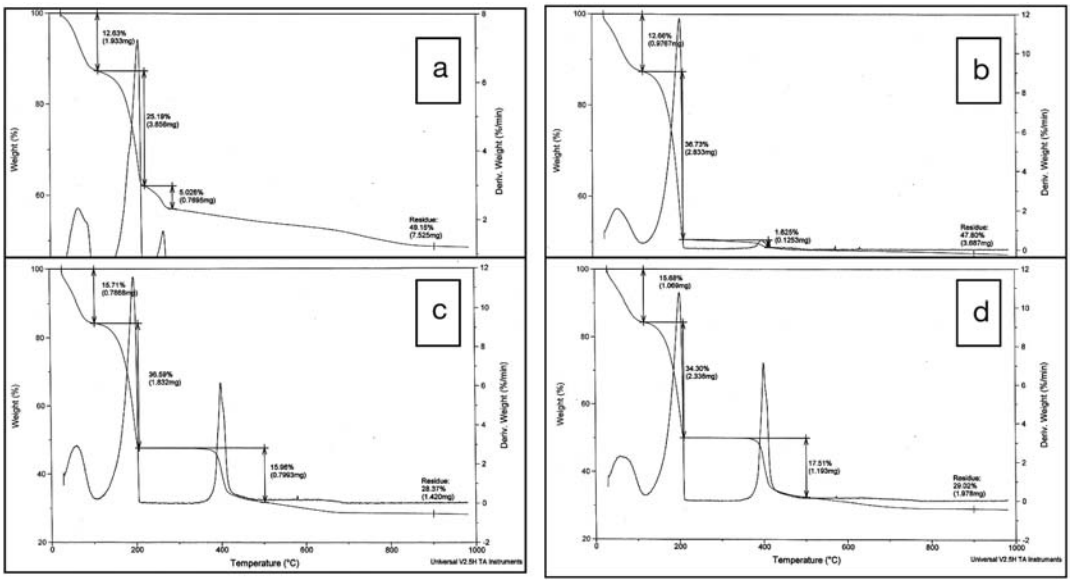

Figure 9. The thermogravimetric analysis and differential thermal analysis (TGA-DTA) thermograms of zinc glycerolate that had been heated in (a) 10%, (b) 30%, (c) 50% and (d) 70% (w/w) glycerol in aqueous solution at 80°C for 2 hr.

In order to establish the optimum concentration of glycerol in aqueous solution that would minimise hydrolysis of its zinc complex, infusions of zinc glycerolate, glycerol and water were prepared, stored at room temperature and characterised after three months. The amount of glycerol was varied from 10% to 100%, while zinc glycerolate was maintained at 10% w/w concentration. The mixture was shaken at 24-hr intervals and later filtered. The filtered particulates were analysed by TGA-DTA to determine zinc glycerolate and zinc oxide contents. A graph was subsequently plotted to reveal the hydrolysis profile of zinc glycerolate in aqueous solution. A replicate set of infusions was prepared but they were subjected to heating at 80°C for 2 hr, filtered and then analysed by TGA-DTA analysis.

The TGA-DTA thermograms of zinc glycerolate that had been heated in various concentrations of glycerol in aqueous solution at 80°C for 2 hr are shown in *Figure 9*. The peak height for glycerolate ion content degradation at 400°C increased substantially with a decrease in glycerol concentration used in the aqueous phase.

The effect of the various amounts of glycerol in aqueous solution on the hydrolysis profile of zinc glycerolate for over three months at room temperature and at 80°C for 2 hr are featured in *Figures 10a* and *10b*, respectively. The decreasing trend of zinc glycerolate in the sample indicated hydrolysis of the complex in aqueous solution. Both figures show that by increasing the amount of glycerol in aqueous solution, hydrolysis of zinc glycerolate complex can be minimized. The demand for glycerol to prevent hydrolysis increased significantly with heating at 80°C for 2 hr as shown in *Figure 10b*. The optimum amount of glycerol needed to minimise the hydrolysis process in aqueous solution was 30% to a maximum concentration of 60% glycerol under high heat and humidity conditions.

The dose response curve of the HIE score on dermal irritection for zinc glycerolate is shown in

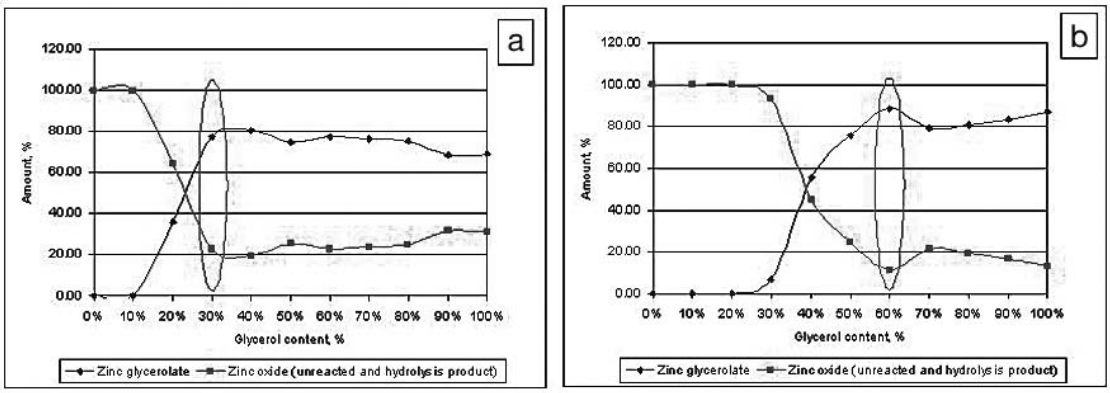

Figure 10. Effect of various amounts of glycerol in aqueous solution on hydrolysis profile of zinc glycerolate (a) for three months at room temperature, and (b) at 80°C for 2 hr.

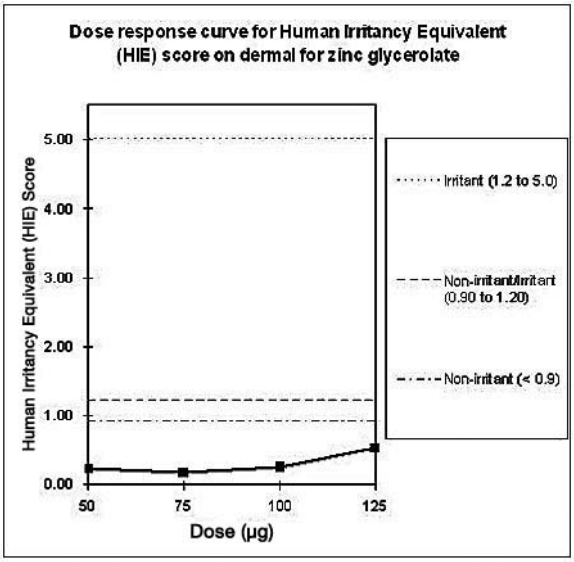

Figure 11. In vitro *dermal irritection score of zinc glycerolate.*

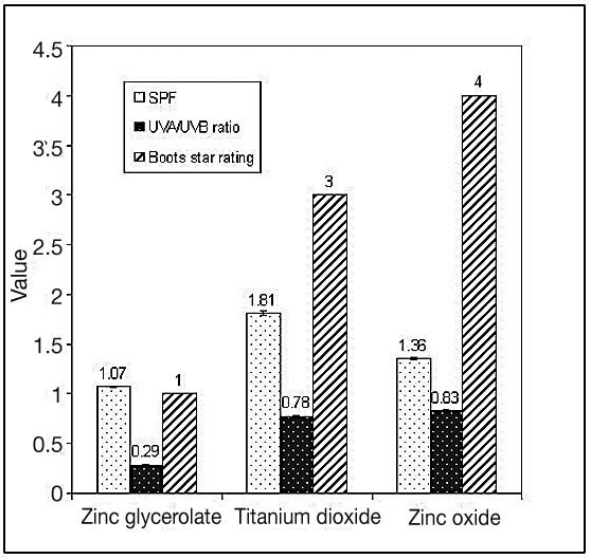

Figure 12. SPF values, UVA to UVB ratios and Boots star rating of zinc glycerolate, titanium dioxide and zinc oxide.

Figure 11, and, based on the results, zinc glycerolate is classified as non-irritant. Therefore, it has good potential to be used in pharmaceutical and personal care products.

Figure 12 shows the SPF values, UVA to UVB ratios and Boots star rating of zinc oxide, titanium dioxide and zinc glycerolate. Zinc glycerolate had the lowest SPF value, UVA to UVB ratio and Boots star rating as compared with zinc oxide and titanium dioxide. The results indicate that zinc glycerolate had a mean SPF of 1.07 ± 0.004 and UVA ratio of 0.29, which are considered moderate (one star) by the Boots star rating for UVA protection. The SPF value of titanium dioxide was 1.81 ± 0.02 and its UVA ratio was 0.78 (three stars), while the SPF value of zinc oxide was 1.36 ± 0.01 and its UVA ratio was 0.83 which is classified as maximum (four stars).

CONCLUSION

The preparation of zinc glycerolate from simple zinc oxide and excess glycerol using microwave technology produced hexagonal prisms of 23.015 µm particle size and having a vastly different appearance from zinc oxide. Characterisation by means of TGA-DTA and X-ray diffraction showed that zinc glycerolate was considered an unstable compound in aqueous condition. Hence, it is recommended that the amount of glycerol to be incorporated in the aqueous phase is 60% to minimise the hydrolysis process. Zinc glycerolate with petrolatum as excipient can be readily used to provide sun protection to the skin with good esthetical quality, because upon application to the skin, it does not produce whitish streaks as are commonly encountered when applying

products using zinc oxide or titanium dioxide. Therefore, zinc glycerolate has great potential to be incorporated into personal care formulations as it is non-irritant to the skin, and is further enhanced by its sun protection properties.

REFERENCES

ANON. (1996). Irritection® Assay System. *Instruction Manual*. In Vitro International, Irvine, CA. p. 1-11.

ANON. SPF analysis of sunscreens using labsphere UV-1000S ultraviolet transmittance analyzer. *Technical Note*. p. 1-12.

APISARIYAKULM, A; BUDDHASUKH, D; APISARIYUKUL, S and TERNAI, B (1990). Zinc monoglycerolate is effective against oral herpetic sores. *The Medical Journal of Australia*, *152*: 54.

BRYLANTS, J and PHILLIPPE, A N (1980). IR and moessbauer study of iron glycerolates. *J. Inorganic and Nuclear Chemistry*, *42 (11)*: 1603-1611.

COULSTON, L and DANSONA, P (1980). Insulin-like effects of Zn^{2+} on adipocytes. *Diabetes*, *29*: 665-667.

FAIRLIE, D P; WHITEHOUSE, M W and TAYLOR, R M (1992). Zinc monoglycerolate – a slow release source of therapeutic zinc: solubilization by endogenous ligands. *Agents and Actions*, *36*: 152-158.

HAMBLEY, T W and SNOW, M R (1983). The crystal and molecular structure of zinc (II) monoglycerolate. *Australian Journal of Chemistry*, *36 (6)*: 1249-1253.

HAZIMAH, A H; BADRI, M; CROUSE, K A and MANAS, A R (2001). Some complexes of glycerol and their applications. *Palm Oil Developments No. 35*: 8-10.

LIDE, D R (1994). *CRC Handbook of Chemistry and Physics*. 75th Edition, Boca Raton, FL, CRC Press. p. 9-79.

MAY, J M and CONTOREGGI, C S (1982). The mechanism of the insulin-like effects of ionic zinc. *J. Biological Chemistry*, *257*: 4362-4368.

MENDOLOVICI, E; SAGARZAZU, A and VILLALBA, R (1986). High temperature synthesis and characteristics of aluminium substituted iron alkoxide. *Thermochimica Acta*, *107*: 75-82.

NARGY, L; BURGER, K; KURTI, J; MOSTAFA, M A; KORECZ, L and KIRICSI, I (1986). Iron (III) complexes of sugar-type ligands. *Inorganica Chimica Acta*, *124 (1)*: 55-59.

PAVIA, D L; LAPMAN, G M and KRIZ, G S (1996). *Introduction to Spectroscopy, A Guide for Students of Organic Chemistry*. 2nd Edition, Saunders College Publishing.

RADOSLOVICH, E W; RAUPACHI, M; SLADE, P G and TAYLOR, R M (1970). Crystalline cobalt, zinc, manganese and iron alkoxides of glycerol. *Australian Journal of Chemistry*, *23*: 1963-1971.

REMIAS, R; KUKOVECZ, A; DARANYI, M; KOZMA, G; VARGA, S; KONYA, Z and KIRICSI, I (2009). Synthesis of zinc glycerolate microstacks from a ZnO nanorod sacrificial template. *European Journal of Inorganic Chemistry*, *24*: 3622-3627.

SHISHEVA, A; GEFEL, D and SHECHTER, Y (1992). Insulin-like effects of zinc ion *in-vitro* and *in-vivo*. *Diabetes*, *41*: 982-989.

SINA, J F; GALER, D M; SUSSMAN, R G; GAUTHERON, P D; SARGENT, E V; LEONG, B; SHAH, P V; CURREN, R D and MILLER, K (1995). A collaborative evaluation of seven alternatives to the Draize eye irritation test suing pharmaceutical imtermediates. *Fund. Appl. Toxicol.*, *26*: 20-31.

TAYLOR, R M (1988). Plastics or rubber materials modified by crystalline glycerato-zinc complex. US patent 4789701.

TAYLOR, R M (1990). Relating to the formation of metal alkoxides and metal powders by the use of microwave radiation. US patent 4943316.

TAYLOR, R M (2006). Microfine zinc-glycerol complexes. US patent application publication. Publication No: US 2006173073A1.

TAYLOR, R M and BROCK, A J (1985). Pharmaceutical compound zinc glycerolate complex prepared by reacting zinc oxide and glycerol. US patent 4544761.

TAYLOR, R M and BROCK, A J (1989). Zn glycerolate complex and additions for pharmaceutical applications. US patent 4876278.

WHITEHOUSE, M W; RAINSFORD, K D; TAYLOR, R M and VERNON-ROBERTS, B (1990). Zinc monoglycerolate: a slow-release source of zinc with anti-arthritic activities. *Agents and Actions*, *31(1-2)*: 47-58.