Review Article

Adverse reactions to cosmetics and methods of testing

P. K. Nigam

The Department of Dermatology and STD, Pt. J. N. M. Medical College, Raipur - 492 001, India

Address for correspondence: Dr. P. K. Nigam, Professor and Head, Department of Dermatology and STD, Pt. J. N. M. Medical College, Raipur - 492 001, C.G, India. E-mail: drpknigam@yahoo.co.in

ABSTRACT

Untoward reactions to cosmetics, toiletries, and topical applications are the commonest single reason for hospital referrals with allergic contact dermatitis. In most cases, these are only mild or transient and most reactions being irritant rather than allergic in nature. Various adverse effects may occur in the form of acute toxicity, percutaneous absorption, skin irritation, eye irritation, skin sensitization and photosensitization, subchronic toxicity, mutagenicity/genotoxicity, and phototoxicity/photoirritation. The safety assessment of a cosmetic product clearly depends upon how it is used, since it determines the amount of substance which may be ingested, inhaled, or absorbed through the skin or mucous membranes. Concentration of ingredients used in the different products is also important. Various test procedures include *in vivo* animal models and *in vitro* models, such as open or closed patch test, *in vivo* skin irritation test, skin corrosivity potential tests (rat skin transcutaneous electrical resistance test, Episkin test), eye irritation tests (*in vivo* eye irritancy test and Draize eye irritancy test), mutagenicity/genotoxicity tests (*in vitro* bacterial reverse mutation test and *in vitro* mammalian cell chromosome aberration test), and phototoxicity/photoirritation test (3T3 neutral red uptake phototoxicity test). Finished cosmetic products are usually tested in small populations to confirm the skin and mucous membrane compatibility, and to assess their cosmetic acceptability.

Key words: Adverse reactions, Cosmetics, Methods of testing

INTRODUCTION

Cosmetics are "articles intended to be rubbed, poured, sprinkled, or sprayed on, introduced into, or otherwise applied to the human body or any part thereof for cleansing, beautifying, promoting attractiveness, or altering the appearance."[1]

A recent study found that an average adult uses nine cosmetic products daily. More than 25% of women use 15 or more.[2] Cosmetics, toiletries, and skin-care products, including sunscreens, quite frequently cause adverse reactions,[3] and are commonest single reason for hospital referrals with allergic contact dermatitis.[4] It is estimated that 1–3% of the population are allergic to a cosmetic or cosmetic ingredient.[5] In one American survey comprising 30,000 consumers, 700 reactions occurred during 1-year period.[6]

From a dermatologist point of interest, cosmetics may be grouped as: (a) skin-care cosmetics (cleansing agents, moisturizing agents, etc.), (b) hair-care cosmetics (shampoos, hair colorants, styling agents, etc.), (c) face-care cosmetics (facial foundations, powders, eye shadows, mascara, lipsticks, etc.), (d) nail-care cosmetics (nail varnishes, paint removers, etc.), (e) fragrance products (deodorants, aftershaves, perfumes, etc.), and (f) ultraviolet (UV) light screening preparations.

Skin cleansing agents remain on the body for a very short period of time and rarely cause significant adverse reactions, however, perfume and others constituents may cause skin irritation and allergic reactions. Moisturizers increase the hygroscopic properties of the skin; however, high concentration of these substances may cause irritation and exfoliation. Skin lightening/depigmenting agents, such as hydroquinone

How to cite this article: Nigam PK. Adverse reactions to cosmetics and methods of testing. Indian J Dermatol Venereol Leprol 2009;75:10-19.

Received: November, 2007. **Accepted:** July, 2008. **Source of Support:** Nil. **Conflict of Interest:** None declared.

(HQ), are one of the most widely prescribed agents, however, with reports of potential mutagenicity and ochronosis, there has been increasing impetus to find alternative herbal and pharmaceutical depigmenting agents, such as kojic acid (5-hydoxy-4-pyran-4-one-2-methyl, 1–4%) and azelaic acid (20% cream). Ochronosis is an uncommon adverse effect of HQ, characterized by progressive darkening of the area to which the cream containing high concentrations of HQ is applied for many years. 'Black henna' tattoo is a chemical stain due to p-phenylenediamine (PPD), in the form of commercial hair dye mixed into the henna paste. Addition of this artificial dye stains the skin in much shorter duration, an hour or less. Adverse reactions to PPD can include stinging sensations, with an erythematous rash, swelling, blisters, and surface oozing. There have been several reports in the literature of immediate allergic (and also anaphylactic) reactions on using henna dyes.[7] Most cases have sneezing, runny nose, cough, and shortness of breath instead of skin reactions. Adverse effects to sun-screening agents may result in irritant, allergic, phototoxic, or photoallergic reactions, and caused not only by the active constituents but also by the additives such as fragrances and stabilizers. Benzophenones are probably the most common sensitizers, while dibenzoylmethanes, para-aminobenzoic acid (PABA), and cinnamates may cause photoallergic dermatitis.[8] The allergic reactions associated with deodorants/antiperspirants and fragrances are usually caused by fragrance or other ingredients. Fragrance can enter the body through lungs, airways, skin, ingestion, and via pathways from the nose directly to the brain and can cause headaches, irritation to eyes, nose, and throat, dizziness, fatigue, forgetfulness, and other symptoms. Fragrance is the number one cause of skin allergic reactions to cosmetics.[9] As much as 15% of the general population may find fragrance a lower airway irritant and as much as 10% of the general population may have skin allergy to fragrance.[9] Fragrance in the air can cause airborne contact dermatitis.[10] Coumarin, methyl eugenol, and others are suspected carcinogens.[8] Some phthalates are suspected hormone disrupters.[11] Shampoos and conditioners have only a brief contact with the skin and are not a common cause of cutaneous irritant or allergic contact dermatitis. However, eye irritation can be a problem. Possible sensitizers in shampoos include formalin, parabens, hexachlorophene, triclosan, and fragrances. Matting of scalp hair is most commonly a sudden, usually irreversible, tangling of scalp hair resulting from shampooing.[12] Hair straightening (relaxing) with pressing oils and heated metal combs or round tongs may be associated with hair-shaft breakage and scarring alopecia.[13] Hair removal techniques may partially account for allergic and photoallergic reactions. The adverse effects of shaving include skin irritation, cuts in the skin, ingrown hair (pseudofolliculitis), etc. The active ingredients in hair bleaches are hydrogen peroxide solutions that oxidize melanin to a lighter color. They may be supplemented with persulfate boosters. The disadvantages of bleaching include skin irritation, temporary skin discoloration, pruritus, and the prominence of bleached hair against tanned or naturally dark skin. Ammonium persulfate may cause types I and IV allergic contact reactions. Also, generalized urticaria, asthma, syncope, and shock in reaction to the persulfate activator have been reported. About 12% of cosmetic reactions occur on the eyelid, mainly due to the eye shadow. Irritant contact dermatitis is more common than allergic contact dermatitis. Mascara is the most commonly used eye cosmetic. The most feared adverse effect of mascaras is that of infection, particularly *Pseudomonas aeruginosa* corneal infections, which can permanently destroy visual acuity, due to multiple reuses of applicator and reinsertions into the tube between uses. Kajal and surma are mainly carbon compounds, but surma also contains mercury or lead and may put at risk of serious health problems. Nail plate discoloration and allergic contact dermatitis are the major dermatological concerns with the use of nail polish. The nail staining is seen more with dissolved rather than suspended pigments.

SAFETY OF A COSMETIC PRODUCT

The adverse reactions may occur to one of the primary constituents of the cosmetic formulation or contamination or procedural misconduct. Preservatives are the second most common cause of skin reactions, besides fragrances. Most reactions being irritant rather than allergic in nature.[14] In most cases, these are only mild or transient such as stinging and smarting, and

Table 1: Evaluation of the safety/toxicity of a cosmetic product

In vitro tests	*In vivo tests*
Screening for severe irritancy	Screening toxicological profile
Phototoxicity	Determination of the no-observed adverse effect levels (NOAEL)
Percutaneous absorption	Adverse effects at higher exposure
Mutagenicity/genotoxicity	

Table 2: Tests for safety/toxicity assessment of a cosmetic product

Test	Testing method
Noninvasive bioengineering techniques	Skin hydration
	Trasepidermal water loss
Patch tests:	1. Open patch tests
	2. Closed patch tests
Photo-patch testing:	For photocontact dermatitis, on exposure to antigen and sunlight.
Screening for fragrance/ perfumes:	TRUE test
Repeated open application test	Provocative Use Test ROAT
Use testing:	Placing cosmetic near the eye for five consecutive nights
Chemical analysis	
Elimination test	
Dimethylgloxime test:	To identify nickel allergy
Tests of irritancy and sensitivity:	1. Soap-chamber tests or use tests
	2. 'Repeat insult' test
	3. The Draize eye irritancy test
Murine Local Lymph Node Assay (LLNA):	For sensitizing potential of a test substance using rabbit's ears but now
Tests of comedogenicity:	on humans are more common
Percutaneous absorption studies	
Tests for skin corrosivity	1. Rat skin 'TER' assay test
	2. Corrositex
	3. Skin 2TM ZK1350 corros.test
	4. Episkin test
Tests of photosensitivity	3T3 NRU PT test
Test for photoallergy	
Test for phototoxicity	
Tests for mutagenicity/genotoxicity:	1. *In vitro* mammalian cell
	2. Chromosome aberration test
Testing of finished cosmetic products:	1. Compatibility test
	2. Acceptability test

contact urticarial.[15] In few cases, reactions may be more severe, with redness, edema, dryness, and scaling. Various adverse effects may occur in the form of acute toxicity, percutaneous absorption, skin irritation, eye irritation, skin sensitization and photosensitization, subchronic toxicity, mutagenicity/genotoxicity, and phototoxicity/photoirritation [Table 1].

SAFETY ASSESSMENT OF A COSMETIC PRODUCT

The safety assessment of a cosmetic product clearly depends upon how it is used, as it determines the amount of substance which may be ingested, inhaled, or absorbed through the skin or mucous membranes. Concentration of ingredients is also important. Too high concentrations result in false-positive reactions, because of their irritant effect, and may even sensitize patients; too low concentrations produce false-negative results [Table 2].[16]

Most of the cosmetic products can be open or closed patch tested as is.[17] Patch testing the individual ingredients separately is preferred. It is advisable to perform open tests before proceeding to closed patch tests, because the effect of irritants is enhanced by occlusion.[18] Shampoos should be diluted to form a 1–2% aqueous solution for closed patch testing and a 5% aqueous solution for open patch testing. Use testing is recommended and performed by placing the eye cosmetic near the eye for five consecutive nights followed by evaluation of the skin for allergic or irritant contact dermatitis. Nail polish can be tested as is. The resin can also be tested alone in 10% petrolatum. Mascaras can be open or closed patch tested as is, but they should be allowed to dry thoroughly prior to closed patch testing to avoid an irritant reaction from the volatile vehicle. For nail-polish removers, only open patch testing, at a concentration of 10% nail polish remover material dissolved in olive oil, should be performed due to its high solvent concentration. For cuticle removers, open patch testing in a 2% aqueous concentration may be used.

Various test procedures including *in vivo* animal models

and *in vitro* models are used to find out the safety level of cosmetic products, such as open or closed patch test, *in vivo* skin irritation test, skin corrosivity potential test (rat skin transcutaneous electrical resistance test, EPISKIN test), eye irritation tests (*in vivo* eye irritancy test and Draize eye irritancy test), mutagenicity/genotoxicity tests (*in vitro* bacterial reverse mutation test and *in vitro* mammalian cell chromosome aberration test), and phototoxicity/photoirritation test (3T3 neutral red uptake phototoxicity test). An important consideration in cosmetic innovation and toxicology is the growing concern about the ethics of testing final/finished products on animals, and it is gradually being discouraged and alternative methods are being designed.

Patch and photo-patch tests

Patch test is useful in knowing the type of reaction to a particular cosmetic, whether irritant or allergic. Also, the standard test series can help in identifying the agents causing allergy. Cosmetics can be classified according to their usage as "leave-on" cosmetics such as lipsticks whose patch test is done "as is".[19] The second variety is the "wash-off" or "rinse-off" cosmetics such as shampoos. They are used in the concentration of 10%. Soaps and detergents are used in the concentration of 1%. To interpret photocontact dermatitis, photopatch test is performed and is considered to be positive, if the test site shows dermatitis on exposure to antigen and sunlight.

Patch test screening for fragrance/perfumes

Balsam of Peru, cinnamal, fragrance mix, and colophony are recognized markers for fragrance allergy. Fragrance mix marketed as TRUE test (Thin-layer Rapid Use Epicutaneous test) contains eight ingredients: eugenol, isoeugenol, oak moss absolute, geraniol, amyl cinnamic aldehyde, hydroxycitronellal, cinnamic alcohol, and α-cinnamal. Fragrance mix detects about 86% of positive reactions. Addition of ylang-ylang oil, narcissus oil, sandalwood oil, and balsam of Peru raised this percentage to 96. Allergy to fragrance can also be tested by using Repeated Open Application Test (ROAT). Balsams, cinnamic alcohol, cinnamaldehyde, benzoic acid, and benzaldehyde can evoke contact urticaria not detected by closed patch testing.

Repeated open application test/provocative use test

In this test,[20] the suspected cosmetic test substance is applied twice daily for up to two weeks to an approximately 5-cm square area on the flexor surface of the forearm near the antecubital area. If no rash appears after one week, the product is considered safe for that individual. This test is applied to screen for allergy to cosmetics including fragrances and to confirm the clinical significance of weak positive patch test reactions.

Thin-layer rapid use epicutaneous test

TRUE test is a reliable allergen patch skin test.[21] The test panels contain 23 different substances or mixes, all of which are well-known causes of contact dermatitis, and a negative control.

Chemical analysis

Occasionally, chemical analysis may be necessary to determine whether a material contains a suspected allergen or to identify new unknown allergens.

Dimethylgloxime test

Dimethylgloxime test is a useful and practical way to identify nickel allergy. It identifies metallic objects that contain enough nickel to provoke allergic dermatitis in individuals allergic to nickel.

Elimination test

Fischer[1] suggested an elimination routine in diagnosis of reactions to cosmetics. All cosmetics are stopped except lipstick, which is allowed if the lips are problem free. When dermatitis has cleared, one cosmetic at a time is tested/allowed. If a reaction occurs, the cosmetic used most recently is eliminated.

Safety testing

The FDA accepts only animal safety data. The most widely used animal test is the 'Draize eye irritancy test' which involves placing drops of the substance in question into the eye of an albino rabbit. The test is positive if any redness, swelling, or cloudiness in the eye is noted.

Tests of irritancy and sensitivity

The irritant potential of a chemical is gauged by: (1) soap-chamber tests or use tests,[22] and (2) 'repeat insult' test.[23]

For irritation potential, a panel size of 12–20 individuals is used. Patches (material) are applied to the skin (of the back usually) with an occlusive dressing and left undisturbed for a 48-hour period. At the end of the 48-hour period, the patch is removed and presence of any reaction is recorded. The substance being tested is then reapplied to the same site occlusively for a further

48-hour period, and this process is repeated three times a week for either a two- or a three-week period. Readings are taken after removal of each patch. This gives the potential for cumulated irritation and will reveal very low orders of toxicity in the preparation. For sensitization potential, a larger panel of at least 100 subjects is taken but a further test is added at the end. After about one week's rest, a further patch of the substance is applied occlusively for a 48-hour period at different site. If after 48 hours a 'positive patch' results, it means that the subject has become sensitized.

The Draize[24] eye irritancy test

Chemical substances (100 mg of a concentrated solution) are dripped into the eyes of six to nine immobilized conscious albino rabbits with their eyes held open with clips at the lid. The progressive damage to the rabbit's eyes is recorded at specific intervals over an average period of 72 hours, with the test sometimes lasting 7–18 days. Reactions to the irritants can include swelling of the eyelid, inflammation of the iris, ulceration, hemorrhaging (bleeding), and blindness. Draize test is considered as crude, imprecise, and not reliable because it is strictly observational and does not adequately reflect the degree of irritancy in humans.

Murine local lymph node assay (LLNA)

LLNA test is applied to test chemicals for allergic contact sensitizing potential of a test substance.[25]

Tests of comedogenicity

These tests[26] were previously conducted using rabbits' ears, but now comedogenicity tests on humans are more common. The test applications are made under occlusion over a period of weeks to the backs or flanks of human volunteer subjects. At the end of this period, either 7-mm-diameter punch biopsies are made or skin surface biopsies (using a cynoacrylate adhesive) are taken. The presence of comedones is assessed microscopically and scored according to number and size.

Toxicity studies

A number of methods are applied to study the toxic behavior of chemicals used in cosmetics. These include both *in vitro* and *in vivo* methodologies. *In vitro* methods have been validated for use in prescreening for severe irritancy, screening of phototoxicity, evaluating the percutaneous absorption, and studying for mutagenicity/genotoxicity. *In vivo* studies are mainly applied to investigate the toxicological profile of a cosmetic ingredient when applied to an animal by a route of exposure (topical, oral, or by the inhalation route) similar to that of human exposure. They allow the determination of the No-Observed Adverse Effect Levels (NOAEL), and also adverse effects at higher exposure.

Percutaneous absorption studies

Percutaneous absorption is defined as the movement of a chemical substance applied to the surface of the skin into the circulatory system. The percutaneous absorbed dose is the amount of a chemical which is systemically distributed. If a substance under investigation is found to have penetrated through the stratum corneum into deeper layers of the skin, it should be considered as having been absorbed. For *in vitro* assessment of percutaneous absorption of cosmetic ingredients, human or pig split-thickness skin is used. The dose as well as the contact time (exposure) with the skin are chosen to mimic intended use conditions. The mass balance of the applied dose and the amounts found in the individual layers of the skin, and on the skin surface are determined. The amounts absorbed are expressed in gm/cm^2 of skin surface and percentage of the applied dose. They are then transformed into mg/kg body weight and thus serve for the assessment of a safety factor.

In vitro tests

Although, human tests are employed as safety assurance tests, but clearly it would be a major advance and much cheaper if such tests could be conveniently performed in the laboratory. There are *in vitro* methods of testing for the whole spectrum of possible adverse effects, but so far only three *in vitro tests* have been scientifically validated: one for phototoxicity and two for skin corrosion. These tests use fragments of human skin and are thus directly applicable to people.

In vitro tests replacing the *in vivo* Draize rabbit test for skin corrosivity

These include:[27] (a) rat skin 'TER' assay test, (b) corrositex, (c) skin 2TM ZK1350 corrosivity test, and (d) Episkin test.

In vitro skin Transcutaneous Electrical Resistance (TER) test has been recommended for the testing of all classes of chemicals. The skin discs are prepared from humanely killed 28–30 days old rats. The test material is applied for up to 24 hours to the epidermal surfaces of skin discs. Corrosive materials are identified by their ability to produce a loss of normal stratum corneum integrity and barrier function, which is measured as

a reduction in the TER below a threshold level. The skin impedance is measured as TER by using a low-voltage, alternating current Wheatstone bridge. In TER corrosivity assay, measurements are recorded in resistance, at a frequency of 100 Hz and using series values. The test substance is considered to be corrosive to skin: (a) if the mean TER value is ≤5 kΩ and the skin disc is obviously damaged, or (b) the mean TER value is ≤5 kΩ, and the skin disc is showing no obvious damage, but the mean disc dye content is greater than or equal to the mean disc dye content of the 10 M HCl positive control obtained concurrently.

Corrositex test did not me*et a*l of the criteria to be considered acceptable as replacement test. The corrosivity potentials of about 40% of the test chemicals could not be assessed with corrositex.

The skin 2 assay had an unacceptably high underprediction rate (57%), although it had a specificity of 100%.

The Episkin test implies a three-dimensional human skin model comprising a reconstructed epidermis with a functional stratum corneum. Test materials are topically applied to the skin for 3, 60, and 240 minutes, and subsequently, assessed for their effects on cell viability by using the MTT assay. The test was able to distinguish between corrosive and noncorrosive chemicals for all of the chemical types.

Tests for mutagenicity/genotoxicity

These include bacterial reverse mutation test (or *in vitro* mammalian cell gene mutation test), and *in vitro* mammalian cell chromosome aberration test.

The bacterial reverse mutation test[28,29] detects chemicals that induce mutations which revert mutations present in the tester strains and restore the functional capability of the bacteria to synthesize an essential amino acid. The bacterial reverse mutation test uses amino-acid-requiring strains of *Salmonella typhimurium* (*S. typhimurium*) and *Escherichia coli* (*E. coli*) to detect point mutations, which involve substitution, addition, or deletion of one or a few DNA base pairs. Test substances are dissolved or suspended in appropriate solvents or vehicles and diluted as appropriate prior to treatment of the bacteria. Concurrent negative (solvent or vehicle) and strain-specific positive controls, both with and without metabolic activation, are included in each assay. Data are presented as the number of revertant colonies per plate.

In vitro mammalian cell chromosome aberration test[30]

This test identifies agents that cause structural chromosome aberrations in cultured mammalian cells. Cell cultures are exposed to the test substance both with and without metabolic activation. At predetermined intervals after exposure of cell cultures to the test substance, they are colchicine treated, harvested, stained, and metaphase cells are analyzed microscopically for the presence of chromosome aberrations with a metaphase-arresting substance such as colcemid. Solid test substances should be dissolved or suspended in appropriate solvents or vehicles prior to treatment of the cells. Liquid test substances may be added directly to the test systems. Proliferating cells are treated with the test substance in the presence and absence of a metabolic activation system. Culture harvest time (cells exposed to the test substance) is 3–6 hours and then cells are subjected to microscopic analysis for chromosome aberrations.

Tests of photosensitivity

Two types of tests are made to test the photosensitizing potential of the test substance: (1) test for phototoxicity and (2) test for photoallergy.

For all phototoxic UV absorbing chemicals, including cosmetic ingredients, routine testing for phototoxicity should be done by an *in vitro* method named "3T3 Neutral Red Uptake Phototoxicity Test (3T3 NRU PT)". Animal models have not been validated for testing phototoxicity.

In vitro 3T3 neutral red uptake phototoxicity test

Compounds that are phototoxic *in vivo* after systemic application and distribution to the skin, as well as compounds that act as photoirritants after topical application to the skin, can be identified by this test.[31,32] The principle of the method is based on a comparison of the cytotoxicity of a chemical when tested in the presence and in the absence of exposure to a noncytotoxic dose of UVA/visible light. This method is based on a cell phototoxicity process, observed in a mammalian cell population *in vitro*. The positive control chemical chlorpromazine is concurrently tested in each assay. A permanent mouse fibroblast cell line, Balb/c 3T3, clone 31, is used. Cytotoxicity in this test is expressed as a concentration dependent reduction of the uptake of the vital dye, neutral red

(NR) 24 hours after treatment with the test chemical and irradiation. Light source emitting UVA and visible regions, both, xenon arcs and (doped) mercury-metal halide arcs are used as solar simulators. A dose of 5 J/cm^2 (UVA) was determined in the validation study to be noncytotoxic to Balb/c 3T3 cells and sufficiently potent to excite even weak phototoxic chemicals. Test procedure is of three days. On the first day, a cell suspension of 1 x 105 cells/ml in culture medium is prepared and 100 μL culture medium is dispensed into the peripheral wells of a 96-well tissue culture microtiter plate. On the second day, cells are incubated with the eight different concentrations of the test chemical in the dark for 60 minutes (7.5% CO2, 37 ºC). On the third day, microscopic evaluation of the cells is done under a phase-contrast microscope and changes in morphology of the cells due to cytotoxic effects of the test chemical are recorded. Results are evaluated as the concentration of the test chemical reflecting a 50% inhibition of the cellular NRU (EC50).

The test has been shown to give excellent predictivity for phototoxicity.[33] The predictive value of this method for a potential human photoxic chemical has been demonstrated to be between 95 and 100%.

Compatibility testing of finished cosmetic products in human volunteers

Since tests in animals and alternative methods are of predictive limited value with respect to human exposure, confirmatory compatibility tests of cosmetic finished products in humans may be needed scientifically and ethically. Finished cosmetic products are usually tested in small populations to confirm the skin and mucous membrane compatibility and to assess their cosmetic acceptability. Two types of tests are applied in human volunteers for the skin compatibility assessment of finished cosmetic products: (1) compatibility test: to confirm that there are no harmful effects when applying a cosmetic product for the first time to the human skin or mucous membrane, and (2) acceptability test: to confirm the fulfillment of the expectations for a cosmetic product in-use.

The European commission scientific committee[34] has issued guidelines on the use of human volunteers in compatibility testing of finished cosmetic products. Children should not be involved with the testing of the compatibility of cosmetic products. Among the most frequently used tests for finished cosmetic products are skin irritation tests as human repeated insult patch tests, chamber scarification tests, ROAT, and soap chamber tests for detergents, and various other occlusive or open test methods developed to simulate intended use situations. Irritancy reaction in humans is not an absolute measure and should be related to appropriate controls defining the range of response. For specific products, confirmatory safety tests may be performed in the surrounding area of the eye. Noninvasive bioengineering techniques such as skin hydration, dry skin, wrinkles, depigmentation, and measurement of trasepidermal water loss (TEWL) can be applied in safety assessment to quantify and objectivate the results, to measure even subclinical symptoms, and to obtain additional information.

Previous methods for skin compatibility testing of colored cosmetics were limited by their ability to detect erythema reactions (reddening) underneath nontransparent products. Recently, a new spectroscopic method to quantify reddening of human skin *in vivo* below colored cosmetics (*e.g.*, hair dye, lipstick, makeup) was developed using spectrophotometer.[35] The skin compatibility of nontransparent cosmetic products was determined by detection of a remission band in the near-infrared spectral region.

Intersensory phenomena frequently occur during the subjective assessment of consumer products and are very difficult to measure properly in an objective way. Previously, the objective emotional assessment (OEA) technique based on the evaluation of psycho–physiological reactions and parameters had proven to be highly suitable for determining emotional consumer response. In a recent study, the intersensory effects of color and fragrance via OEA was assessed and it was found that OEA could be successfully applied to such weak stimuli as color and fragrance and there was a good differentiation of matched and mismatched combinations with respect to their activation and emotional effect on volunteers. A very subtle separation of stimuli was achieved, which allows deep insight into the mutual interdependency of color and fragrance.[36]

INDIAN PERSPECTIVE

Dogra, Minocha and Kaur[37] observed the incidence of contact allergic dermatitis to be 3.3% with various cosmetics used by the patient. The most common type of adverse reaction to cosmetics seen in their

patients was contact allergic dermatitis in 59.2% mainly to hair dyes, shaving creams, and lipsticks. Photoallergic dermatitis was seen in 35%, only to hair dyes and lipsticks. The other less common reactions were contact irritant dermatitis, hyperpigmentation, hypopigmentation, contact urticaria, acneiform eruptions, hair breakage, and nail breakage. Multiple sensitivities were seen with various cosmetics and their ingredients in few cases. PPD is found to be a very strong sensitizer and a common contact allergen in hair dyes, in 35–42% of cases.[37,38] A causal link such as shaving cream with isopropyl myristate and musk mix,[39] soaps with chloroxylenol, [40] jasmine absolute and synthetic, lipsticks with propyl gallate,[41] bindis with tertiary butyl hydroquinone,[42] and face cream with bronopol, butyl hydroxy anisole, cetyl alcohol, isopropyl myristate, sorbitan mono-oleate, sorbitan sesquioleate, triethanolamine, and various perfumes, etc. has been shown. After-shave lotions mainly contain alcohol, aluminium chlorohydroxide, menthol, camphor, and glycerine. Contact dermatitis to shaving preparations is mainly due to after-shave lotions and perfume. Patch testing with shaving cream is done either with the finished product or with individual ingredients.[43,44] Contact lenses are widely used both for cosmetic and therapeutic purposes. Fernandez[45] has emphasized the various complications like bacterial and fungal infections, damage to the epithelium, substantia propria, and even the endothelium, and have discussed better methods of sterilization and better fitting to reduce the complications They also suggested that the material used for manufacturing contact lens should be resistant to infection, easy to clean, and have good oxygen permeability. Hans *et al*,[46] assessed the phototoxic potential of cosmetic products and found some of the lipsticks and facial creams generated reactive oxygen species (ROS), produced hemolysis, and caused lipid peroxidation in human erythrocytes (*in vitro*) when exposed to sunlight. The test lipsticks and creams showed absorption in UV/visible range. The study demonstrated synergistic action of cosmetic products and sunlight, and thus, suggested that sunlight exposure should be avoided after the use of photosensitive cosmetics. Bhargava and Mathew[47] have recently reported a case of hair dye poisoning, mainly due to combined toxicities of sodium EDTA and PPD. Chanchal and Swarnalata[48] have described the various novel approaches in herbal cosmetics which could improve both the esthetic appeal and performance of a cosmetic product. In this respect, the approaches studied and discussed include liposomes, phytosomes, transferosomes, nanoemulsions, nanoparticles, microemulsions, nanocrystals, and cubosomes. Dogra and Dua[49] emphasized the main problem in cosmetic dermatitis is to identify the allergen as number of agents are being used by the patients. Also, in India, there is no legislation regarding labeling on the cosmetics as in West; so no clear-cut information regarding ingredients is available. In India, the 'Drugs and Cosmetics Act'[50] is mainly aimed to regulate the import, manufacture, distribution, and sale of drugs and cosmetics. The central or state government have power to make rules and appoint inspector to control or inspect any drug or cosmetic for its standardization and safety which can be tested in the central or state drug laboratory. The government can prohibit manufacturing, importing, or selling of any drug or cosmetic. Violation of law by any person or corporate manager or owner is liable for punishment for a term which may extend to 3–10 years and shall also be liable to fine which could be five-hundred or ten-thousand rupees or with both. Drugs and cosmetic rules 1995 contains the list of drugs for which license is required by manufacturer, importers, and exporters.

CONCLUSION

Although, cosmetic products have rarely been associated with serious health hazards, this does not mean that cosmetics are always safe to use, especially with regard to possible long-term effects as the products may be used extensively over a large part of the human lifespan. Cosmetics and personal-care products may contain ingredients whose safety is unclear or which are known to pose health risks. Testing of cosmetic products is voluntary and controlled by manufacturers. Many of the cosmetics, primarily the hair dyes and shampoos may contain ingredients classified as known or probable human carcinogens. Furthermore, many of them may also contain "penetration enhancers" increasing penetration through the skin. Little research is available to document the safety or health risks of low-dose repeated exposures to chemical mixtures like those used in personal-care products and the absence of data should never be mistaken for proof of safety.

REFERENCES

1. Fischer AA. Cutaneous reactions to cosmetics. 2nd ed. Philadelphia: Lea and Febiger; 1973. p. 217-41.
2. Linda B, Sedlewicz BS. Cosmetic preservatives: Friend or foe? Skinmed 2005;4:8-100.
3. Foley P, Nixon R, Marks R. Frowen K, Thompson S. The

frequency of reactions to sunscreens: Results of a longitudinal population based study on the regular use of sunscreen in Australia. Br J Dermatol 1993;128:512-8.
4. de Groot AC. Labelling cosmetics with their ingredients. Br Med J 1990;300:1636-8.
5. De Groot AC, Beverdam EG, Tjong Ayong Ch, Coenraads PJ, Nater JP. The role of contact allergy in the spectum of adverse effects caused by cosmetics and toiletries. Arch Dermatol 1988;124:1525-9.
6. Grief M, Maibach HI. Cosmetic ingredient labelling. Contact Dermatitis 1977;3:94-7.
7. Nigam PK, Saxena AK. Allergic contact dermatitis from henna. Contact Dermatitis 1988;l8:55-6.
8. Johansen JD, Rastogi SC, Menne T.Johansen JD. Threshold responses in cinnamic-aldehyde-sensitive subjects: Results and methodological aspects. Contact Dermatitis 1996;34:165-71.
9. Schafer T, Bohler E, Ruhdorfer S, Weigl L, Wessner D, Filipiak B, *et al*. Epidemiology of contact allergy in adults. Allergy 2001;56:1192-6.
10. de Groot AC, Frosch PJ. Adverse reactions to fragrances: A clinical review. Contact Dermatitis 1997;36:57-86.
11. Bridges B. Fragrance: emerging health and environmental concerns. Flavour Fragrances J 2002;17:361-71.
12. Wilson CL, Ferguson DJ, Dawber RP. Matting of scalp hair during shampooing: A new look. Clin Exp Dermatol 1990;15:139-42.
13. LoPresti P, Papa CM, Kligman AM. Hot comb alopecia. Arch Dermatol 1968;98:234-8.
14. Eiermann HJ, Larsen W, Maibach HI, Taylor JS. Prospective study of cosmetic reactions: 1977-1980. J Am Acad Dermatol 1982;6:909-17.
15. Maibach HI, Johnson HL. Contact urticaria syndrome. Arch Dermatol 1975;111:726-30.
16. Fisher AA. Cosmetic actions and reactions: Therapeutic irritant, and allergic. Cutis 1980;26:22-9.
17. de Groot AC. Contact allergy to cosmetics: Causative ingredients. Contact Dermatitis 1987;17:26-34.
18. Magnusson B, Hersle K. Patch test methods: Influence of adhesive tape on test response. Acta Derm Venereol 1966;46:275-8.
19. Bourke J, Coulson I, English J, British association of dermatologists: Guidelines for care of contact dermatitis. Br J Dermatol 2001;145:877-85.
20. Hannuksela M, Salo H. The repeated open application test (ROAT). Contact Dermatitis 1986;14:221-7.
21. Kist JM, el-Azhary RA, Hentz JG, Yiannias JA. The contact allergen replacement database and treatment of allergic contact dermatitis. Arch Dermatol 2004;140:1448-50.
22. Frosch PJ, Kligman AM. The soap chamber test: A new method for assessing the irritancy of soaps. J Am Acad Dermatol 1979;1:35-41.
23. Farage MA, Bjerke DL, Mahony C, Blackburn KL, Gerberick GF. A modified human repeat insult patch test for extended mucosal tissue exposure. Contact Dermatitis 2003;49:214-5.
24. Draize JH, Woodard G, Calvery HO. Methods for the study of irritation and toxicity of substances applied topically to the skin and mucous membranes. J Pharmacol Exp Ther 1944;82:377-90.
25. Kimber I, C Weisenberger. A murine local lymph node assay for the identification of contact allergens: Assay development and results of an initial validation study. Arch Toxicol 1989;63:274-82.
26. Simion FA. Acnegenicity and comedogenicity testing for cosmetics. In: Barel AO, Paye M, Maibach HI, editors. Handbook of Cosmetic Science and Technology. New York: Marcel Dekker, Inc; 2001. p. 837-44.
27. Europian Commission: Notes of guidance of cosmetic ingredients for their safety evaluation, 1999. Available from: http://www.leffingwell.com/cosmetics/vol_3en.pdf.
28. Ames BN, Mccann J, Yamasaki E. Methods for detecting carcinogens and mutagens with the Salmonella/mammalian microsome mutagenicity test. Mutat Res 1975;31:347-64.
29. Green MHL, Muriel WJ. Mutagen testing using trp+ reversion in *Escherichia coli*. Mutat Res 1976;38:3-32.
30. FDA recommendations for conducting an *in vitro* Mammalian Chromosomal Aberration Test [updated on 2003 Nov]. Available from: http://www.cfsan.fda.gov/~redbook/ redivc1b. html.
31. Spielmann H, Balls M, Dupuis J, Pape WJ, Pechovitch G, deSilva O, *et al*. The international EU/COLIPA *in vitro* phototoxicityvalidation study: Results of phase II (blind trial): The3t3 NRU phototoxicity test. Toxicol Invitro 1998;12:305-27.
32. European Commission Statement on the Scientific Validity of the 3T3 NRU PT Test (an *in vitro* test for phototoxic potential). Ispra, Italy: Joint Research Centre, Environment Institute; 1997.
33. Zhang HW, Dong SX. Study on using *in vitro* 3T3 neutral red uptake to test phototoxicity. Wei Sheng Yan Jiu 2005;34:628-30.
34. European commission scientific committee guidelines on the use of human volunteers in compatibility testing of finished cosmetic products. Available from: http://ec.europa.eu/health/ph_risk/committees/sccp/docshtml/sccp_out87_en.htm
35. Rohr M, Ernst N, Schrader A. Compatibility testing of colored cosmetics: A new tool for objective testing near infrared remission spectroscopy (NIR-RS). Internat J Cosmetic Sc 2005;27:291.
36. Eisfeld W, Schaefer F, Boucsein W, Stolz C. Tracking intersensory properties of cosmetic products via psycho-physiological assessment. Internat J Cosmetic Sc 2005;27: 292-7.
37. Dogra A, Minocha YC, Kaur S. Adverse reactions to cosmetics. Indian J Dermatol Venereol Leprol 2003;69:165-7.
38. Pasricha JS. Contact dermatitis in India, general features. New Delhi: The off-setters; 1988. p. 1-20.
39. Calnon CD. Quinazoline yellow SS in cosmetics. Contact Dermatitis 1976;2:160-6.
40. dc Groot AC, Weyland JW. Kathon CG: A review. J Am Acad Dermatol 1988;18:350-8.
41. Bajaj AK, Gupta SC, Chatterjee AK, Singh KG, Basu S, Kant A. Hair dye depigmentation. Contact Dermatitis 1996;35:56-7.
42. Fisher AA, Dooms-Goosens A. Persulfate hair bleach reactions. Arch Dermatol 1976;111:1407-9.
43. Mehta SS, Reddy BS. Cosmetic dermatitis: Current perspectives. Int J Dermatol 2003;42:533-42.
44. Pasricha JS. Contact Dermatitis caused by shaving creams. Contact Dermatitis 1988;67-85.
45. Fernandez ST. Contact lenses-Are they safe enough? Indian J Ophthalmol 1987;35:59-61.
46. Hans RK, Agrawal N, Verma K, Misra RB, Ray RS, Farooq M. Assessment of the phototoxic potential of cosmetic products. Food Chem Toxicol 2008;46:1653-8.
47. Bhargava P, Mathew P. Hair dye poisoning. J Assoc Physicians India 2007;55:871-2.
48. Chanchal D, Swarnlata S. Novel approaches in herbal cosmetics. J Cosmet Dermatol 2008;7:89-95.
49. Dogra A, Dua A. Cosmetic dermatitis. Indian J Dermatol 2005;50:191-5.
50. The Drugs and Cosmetic Act, 1940; *The Gazette of India. The Drugs and Cosmetics Act 1940. No. 23 of 1940.Drugs and Cosmetic Rules 1995 vide Gazette Notification - GSR No. 86 (E) A 6/2/02. Available from:* http://www.nihfw.org/ndc-nihfw/ html/Legislations/ The Drugs And Cosmetics Act.htm.

Multiple Choice Questions:

1. Most of the skin allergic reactions to cosmetics are caused by:
   a. Vehicle
   b. Fragrance
   c. Color chemicals
   d. Detergents
2. The adverse effects to cosmetics may occur in the form of:
   a. Acute toxicity
   b. Photosensitization
   c. Mutagenicity/genotoxicity
   d. Any of the above
3. The test used to detect the irritancy of a chemical is:
   a. The Draize eye test
   b. ROAT test
   c. Dimethylgloxime test
   d. Corneometry
4. Which of the following is not an *in vitro* test replacing the in vivo Draize rabbit test for skin corrosivity:
   a. Rat skin 'TER' assay test
   b. Corrositex
   c. Mammalian cell chromosome aberration test
   d. Episkin test
5. For patch testing, "wash-off" or "rinse-off" cosmetics, such as shampoos are used in a concentration of:
   a. 10%
   b. 25%
   c. 40%
   d. 50%
6. Transcutaneous Electrical Resistance (TER) test is a type of:
   a. Patch test
   b. Repeated open application test
   c. TRUE test
   d. Corrosivity test
7. Tests of irritancy include:
   a. Soap-chamber tests
   b. 'Repeat insult' test
   c. Draize test
   d. All of the above
8. *In vitro* tests replacing the in vivo Draize rabbit test for skin corrosivity include:
   a. Rat skin 'TER' assay test
   b. Corrositex
   c. Episkin test
   d. All of the above
9. Tests for mutagenicity/genotoxicity include:
   a. Bacterial reverse mutation test
   b. *In vitro* mammalian cell chromosome aberration test
   c. Both
   d. None
10. Phototoxic and photoirritants potential of cosmetics can be tested by:
    a. 3T3 neutral red uptake phototoxicity test'
    b. Rat skin 'TER' assay test
    c. Draize test
    d. Dimethylgloxime test

Answers
1-b 2-d 3-a 4-c 5-a 6-d 7-d 8-d 9-c 10-a