Solar Radiation and Phototoxicity of Cosmetics: Avenues of In Vitro Skin Models

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Mohd. Danish Kamar, Madhu Bala, Apeksha Vikram, Sunil Kumar Patel, Gaurav Prajapati, Vinay Shakti Shukla, and Saroj Kumar Amar

Abstract

Skin is the body's largest organ, performing several critical functions, including photoprotection and thermoregulation. The skin comprises three distinct layers, each having different types of cells. The epidermis, the outermost layer of skin, contains melanin—a photoprotective pigment to protect from the hazardous effect of UV radiation. Keratinocytes and melanocytes are the primary cell type in the epidermis and provide the barrier against UV radiation and pathogens. Fibroblasts are the major connective tissue in the dermis region that is essential in regulating skin physiology and wound healing. Skin is a protective tool against the external environment, getting exposed to solar radiation, environmental contaminants, and the chemical ingredients of skin care products. Some chemical elements of cosmetics are photosensitive and cause phototoxicity under ambient sunlight. Photosensitized chemicals damage various cell organelles directly or indirectly through photosensitizer-induced oxidative damage. Several models have been developed to study the phototoxicity potential of chemicals/ingredients

M. D. Kamar · M. Bala · A. Vikram · S. K. Patel · G. Prajapati
Photobiology Laboratory, Systems Toxicology & Health Risk Assessment Group, CSIR-Indian Institute of Toxicology Research (CSIR-IITR), Lucknow, Uttar Pradesh, India

Academy of Scientific and Innovative Research, AcSIR Headquarters, CSIR-HRDC Campus, Ghaziabad, Uttar Pradesh, India

V. S. Shukla
Fragrance & Flavour Development Centre, Kanauji, Uttar Pradesh, India

S. K. Amar (✉)
Photobiology Laboratory, Systems Toxicology & Health Risk Assessment Group, CSIR-Indian Institute of Toxicology Research (CSIR-IITR), Lucknow, Uttar Pradesh, India

Environmental Lab, US Army Engineer Research and Development Center, Vicksburg, MS, USA

School of Bioscience and Bioengineering, Lovely Professional University, Phagwara, Punjab, India

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of cosmetic formulation. 2D skin models contain only one cell type and are widely used by researchers. 3D skin models mimic the physiology of human skin by having multiple cell types that are layered like actual skin. 2D skin models are cost-efficient and easy to maintain. At the same time, the 3D skin models represent the environment of the in vivo processes and also can be used to study barrier penetration of cosmetics ingredients. As per a previous study and OECD recommendation, the dermal phototoxicity potential of cosmetics ingredients in humans can be effectively assessed by in vitro approaches.

Keywords

Phototoxicity · Solar radiation · 3D skin models · Cosmetics · UV radiation

Abbreviations

ECM	Extracellular matrix
IC50	concentration at which 50% decrease in cell viability
IL-1α	Interleukin 1 alpha
LDH	Lactate dehydrogenase
MPE	Mean photo-effect
MTT	3-(4, 5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide
NHEK	Normal, human-derived epidermal keratinocytes
NRU	Neutral red uptake
OECD	Organisation for Economic Cooperation and Development
PIF	Photo-irritation factor
RHE	Reconstructed human epidermis
ROS	Reactive oxygen species
UVR	Ultraviolet radiation

6.1 Introduction

In sunlight/UV exposure, some cosmetic formulations applied topically or systemically may lead to phototoxic skin irritation. Cosmetics are generally applied onto the skin, and systemic exposure occurs through percutaneous absorption. When a photoreactive chemical gets excited by absorbing UV or visible light, it becomes phototoxic (Lee et al. 2017). The cosmetic formulation contains numerous chemicals, and the safety evaluation of these cosmetic chemicals was previously done by oral or topical exposure of various formulations on animals for toxicity testing (Vinardell and Mitjans 2017). However, the EU has banned the use of animals for testing and safety evaluation of cosmetics (Couteau and Coiffard 2010). The in vitro assays have been developed and validated by the Organization for Economic Cooperation and Development (OECD). The OECD has provided test

guidelines for in vitro skin testing (OECD 2019). The main cosmetic ingredients related to light absorption and skin irritation are hair dyes, preservatives, sunscreen UV filters, etc. (Mujtaba et al. 2021). Over the years, many models have been developed for the in vitro testing of chemicals. Researchers have widely used 2D models for phototoxicity testing. However, 2D models do not represent the actual human skin. To better mimic the human skin condition, 3D skin cell culture was developed, which contains all the skin layers and the significant skin cell types. To better understand the phototoxicity of cosmetics and its testing, this chapter aims to explain 2D and 3D models for phototoxicity and the advantages and disadvantages of each model.

6.2 Solar Radiation

Solar radiation is optical radiation that comprises a range of radiation such as infrared radiation (IR), visible light, and ultraviolet radiation (UVR). Even though longer wavelengths (radiofrequency and microwave) and shorter wavelengths, both radiations are present. The life of terrestrial organisms mainly depends on the radiation energy generated by the sun. UV radiation wavelength lies in the range of 100–400 nm, and this UV range is further subdivided into the other group of radiation based on wavelengths such as UVA (320–400 nm), UVB (290–320 nm), and UVC (100–290 nm) (de Paula Corrêa 2015). Total UV radiation contains 95% of UVA and 5% UVB, and UVC does not reach the earth's surface due to stratospheric ozone. Initially, the sun was the only source of exposure, but as advancements emerged in science, some artificial sources were present for UV exposure. Due to the advent of this artificial source of radiation, opportunity for exposure to radiation has increased. Frequently repeated exposure to UV radiation can cause skin cancer because ultraviolet radiation that damages the skin is a carcinogenic factor in sunlight (de Paula Corrêa 2015). The accumulation of mutation induced by UV damage is the leading cause of skin cancer initiation. The abundance of UV radiation in the environment contributes to various skin illnesses, including skin cancer, aging, and inflammation. Initially, humans were mainly exposed to UV radiation through the occupational source of sunlight (Pfeifer 2021). Recently, UV exposure has increased due to outdoor activities and tan cosmetics as an artificial source of UV advent. Photons of UV fall under the wavelength of visible light and the wavelength of gamma radiation because UV radiation is a component of the electromagnetic spectrum (Malis et al. 2007). Based on electro-physical properties, the physical energy of UV radiation is divided into UVA, UVB, and UVC (Gallagher et al. 2014). As there is an inverse relationship between wavelength and energy, UVC radiation has the shortest wavelength (100–290 nm) with the highest energy and UVA has the longest wavelength (320–400 nm) but the least energy. At the same time, UVB lies in between UVA and UVC radiation. UV radiations affect molecules, cells, and tissues (D'Orazio et al. 2014). UV exposure varies according to geographical location and sunlight

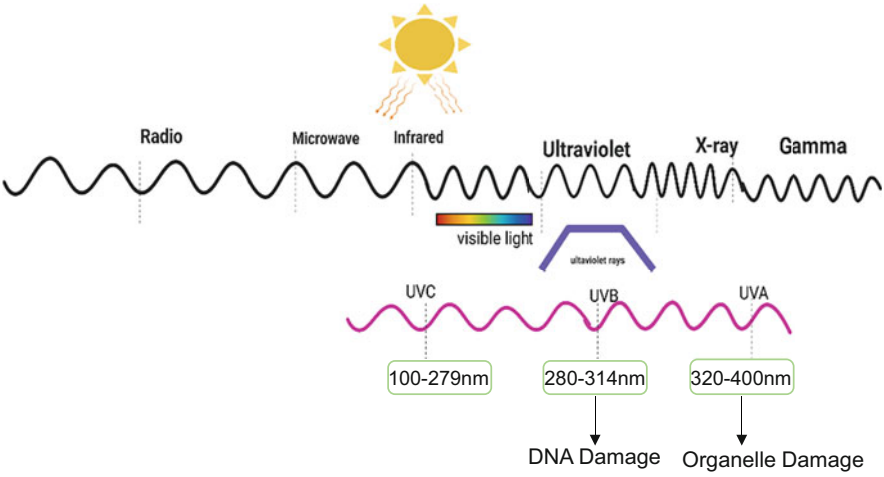

Fig. 6.1 Electromagnetic radiation spectrum showing types of UV, i.e., UVA, UVB, and UVC and the damage on cell

intensity because the sun's intensity varies at a particular spot on the planet (Fig. 6.1).

6.3 Skin Physiology

Skin is the largest organ of the body accounting for 15% of total body weight. It performs several vital functions, including thermoregulation, providing a physical barrier against pathogens, synthesizing vitamin D, and preventing the body from excessive water loss (Zouboulis and Makrantonaki 2011).

The skin is made of three layers: epidermis, dermis, and hypodermis. Each layer has a different structure and cellular composition. The outermost layer, the epidermis, mainly contains keratinocyte cells, which synthesize keratin protein with a protective role. The middle layer, the dermis, constitutes collagen, a fibrillar protein that lies on the panniculus or subcutaneous tissue consisting of lobes of fat cells called lipocytes. The thickness of each layer depends upon the body region, such as the palm and soles having the thickest epidermis and the eyelid having the thinnest epidermis layer (Baroni et al. 2012).

6.3.1 Epidermis

The epidermis is a stratified epithelium layer mainly composed of keratinocytes and dendritic cells and also consists of other cells like Langerhans cells, melanocytes, and Merkel cells. The epidermis is divided into several structural and functional layers, stratum germinativum (basal cell layer), stratum (squamous cell layer),

stratum granulosum (granular cell layer), and stratum corneum (horny or cornified cell layer) (Lai-Cheong and McGrath 2017).

Keratinocytes: Around 80% of cells in the epidermis are keratinocytes. Their appearance changes from one layer to another. It starts from the basal layer of the epidermis and migrates upward in the skin through the differentiation process, keratinization, in which keratinocytes go through a synthetic and degradative phase. The basal layer contains column-shaped keratinocytes that are attached to the basement membrane. Basal cells are clonogenic and can get affected by mutagenic chemicals, affecting the rate of cell division and cell proliferation machinery. Above the basal layer resides a 5–10 cell thick squamous cell layer, spinous cells differ in shape and structure based on their location. Basal spinous cells are polyhedral shape, while the upper layer cells become flattened as they are pushed upward (Choi and Lee 2015). The granular layer is composed of cells with a high amount of keratohyalin granules in the cytoplasm that are required for synthesizing and modifying proteins responsible for keratinization. The thickness of the granular layer depends upon overlying cornified cells. Above the fine layer are the horny cells that provide mechanical protection to underlying cells and a barrier against pathogens to prevent water loss. The cells are flattened and have lost their nuclei during terminal differentiation. It takes 14 days to reach the cell from the basal layer to the topmost layer. Keratinocytes protect against invading pathogens such as microbes, viruses, and fungi from UV radiation and minimize solute, heat, and water loss (Massoud and Rezaei 2014).

Melanocytes: Melanocytes are pigment-synthesizing cells originating from the neural crest cells and mainly located in the basal layer. Melanocytes produce melanin that gives color to the epidermis, hair, and iris. Melanocytes have a round membrane-bound organelle that synthesizes melanin, known as melanosome, and transfer it to keratinocytes (Cichorek et al. 2013). In white skin, melanosomes tend to be released slower in keratinocytes than in dark skin and the rate of degradation of melanosomes compared to dark skin. UV radiation increases the process of melanogenesis and transfer of melanosomes to keratinocytes, leading to skin tanning (Abdel-Malek et al. 2010).

Langerhans cells are derived from bone marrow and migrate to the epidermis during embryonic development. Langerhans cells are members of the dendritic cell/macrophage family. They are specialized to sense the environment and interpret the microenvironmental context to generate an appropriate immune response (inflammation or tolerance). Langerhans cells account for 2–8% of the total cell population in the epidermis (Otsuka et al. 2018).

6.3.2 Dermis

The dermis is a connective tissue layer that gives the skin elasticity and strength. The dermis is rich in nerves and vascular networks and has mast cells, macrophages, and leukocytes alongside the fibroblast cells.

The dermis sits between the two layers, the epidermis and subcutaneous tissue. It interacts with the epidermis at a dermo-epithelial junction that repairs and remodels the skin during wound healing. The dermis contains two layers: reticular dermis (bottom layer) and the papillary dermis (top layer). The reticular dermis is thick and contains blood vessels, nerves, glands, and fat cells. A network of elastin and collagen fibers surrounds the reticular dermis and provides skin with its overall structure and elasticity. The papillary dermis is thinner and contains fibroblast cells, fat cells, touch receptors, macrophages, and blood vessels. The papillary dermis interlocks with the basement membrane of the epidermis. The primary function of the dermis is to provide strength and elasticity, and the blood vessels transport nutrients to the epidermis. The nerve endings allow us to feel pain, pressure, heat, cold, and itchiness (Rippa et al. 2019).

Fibroblast: Fibroblasts are the most abundant cells in connective tissues. They contribute to the secretion of the extracellular matrix that maintains the structural integrity of connective tissues. Fibroblasts produce various products, such as collagen, proteoglycans, laminins, metalloproteinases, and fibronectin. Fibroblast regulates skin physiology and wound repair (Driskell and Watt 2015).

6.3.3 Hypodermis

The innermost layer of the skin is also known as the subcutaneous layer. Hypodermis sits directly beneath the dermis layer and above the other tissue, such as muscle and bone. Fibrous septa separate fat lobules or lipocytes. Hypodermis provides insulation, support, mechanical integrity, buoyancy, and as a storehouse of energy. Hypodermis produces a hormone called leptin, which helps regulate body weight through the hypothalamus (Gilaberte et al. 2016) (Fig. 6.2).

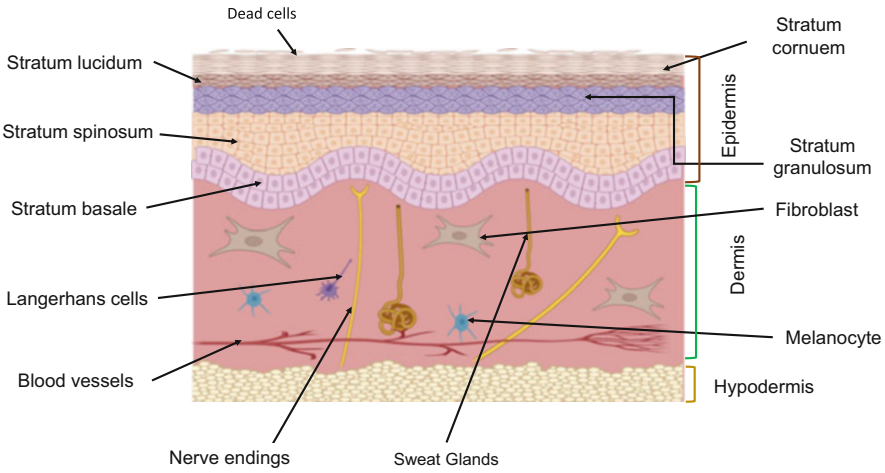

Fig. 6.2 Schematic view of human skin showing different layers of skin (Xiao et al. 2021)

6.4 Phototoxicity

Phototoxicity is defined as "*a toxic response from a substance applied to the body which is either elicited or increased (apparent at lower dose levels) after subsequent exposure to light, or that is induced by skin irradiation after systemic administration of a substance*" (Guidelines 2019). Skin is the largest organ with the body's surface area and is regularly exposed to several man-made products and environmental pollutants. Phototoxicity develops when chemicals come in contact with the skin and get activated by the sunlight, forming cytotoxic products in the skin cells. Skin irritation, erythema, and edema are phototoxicity symptoms similar to the symptoms of exaggerated sunburn. A variety of chemicals have the potential to induce phototoxicity because their structure probably contains benzene rings and heterocyclic rings that can absorb sunrays (Glatz and Hofbauer 2012).

6.4.1 Mechanism of Phototoxicity

A molecule/compound (photosensitizer) needs to absorb photons for a phototoxic reaction to occur. After absorbing the photon, the molecule promotes from its ground state to an excited state. This is the singlet/triplet state of the photosensitizer. The singlet/triplet state has higher energy than the ground state and depends on the spin state of the two electrons with the highest energy. When the two electrons have opposite spin, it is the singlet state, and when the electrons have the same spin, it is the triplet state. This singlet/triplet excited state is only stable for a short time; the triplet state is steady for 10–6 s, and the singlet state is stable only for 10–10 s. When the molecule returns from the excited state to the ground state, the energy absorbed is released by the emission of radiation, heat, or the formation of a photoproduct (chemical reaction) (Baier et al. 2006).

The mechanism of phototoxicity can be divided into direct and indirect modes of action. The direct mode of phototoxicity arises when a chemical absorbs light and gets converted into an excited state, and combines with a crucial cell constituent or transfers electron or hydrogen atoms. This transfer may convert into a toxic radical that may be toxic in subsequent reactions. At the same time, an indirect mode of phototoxicity arises when a chemical absorbs light, goes from an excited singlet state to a triplet state, and reacts with molecular oxygen to generate singlet excited oxygen. Sometimes, a chemical gets excited and transfers an electron to oxygen to create a superoxide anion (Kim et al. 2021).

The exact target of the cell's phototoxic reaction depends on the phototoxic agent's physical and chemical properties. Cosmetics applied topically are more likely to damage the keratinocytes in the epidermis layer, and drugs that are used systemically cause phototoxicity to the dermis layer (Glatz and Hofbauer 2012). The phototoxic reaction can damage cellular organelles. Lipophilic photosensitizers can quickly diffuse in the cell and destroy many cell organelles, such as mitochondria, nuclei, or lysosomes. Hydrophilic photosensitizers can damage the lipid bilayer. Direct damage to cell organelles causes the release of mediators such as histamine

and eicosanoids and initiates the inflammatory response. Some cosmetic ingredients exposed under UVR lead to reactive oxygen species (ROS) generation, which causes the oxidation of cellular organelles and cell membranes. Cosmetics such as lipsticks, creams, lotions, and hair dye have different levels of toxicity due to having specific absorption spectra. P-phenylenediamine, a hair dye ingredient that is photosensitized, leads to apoptosis due to lysosome and mitochondria dysfunction (Goyal et al. 2015). Methylparaben, a preservative used in cosmetics when photosensitized, generates ROS and leads to cell death. Skin inflammation disease occurs when these photosensitized chemicals are used in excess (Dubey et al. 2017).

OECD officially approves 3 T3 neutral red uptake assay for phototoxicity testing. The test evaluates photocytotoxicity by determining the cell viability reduction after exposure to the test chemical in the absence and presence of UV/visible light irradiation. Criteria for any chemical for conducting a 3 T3 NRU test is that the chemical shows absorption in UV/visible region and dissolved in an appropriate solvent. Neutral red dye is a weak cationic dye that quickly penetrates the cell membrane and accumulates intracellularly in the lysosomes of viable cells. To check the phototoxicity potential of the test chemical, the photo-irritation factor (PIF) or mean photo-effect (MPE) is calculated. Photo-irritation factor is the ratio of IC50 (concentration at 50% decrease in cell viability) of non-radiated test chemical over irradiated test chemical. The mean photo-effect is the difference between the dark and light curves at arbitrary doses. PIF value less than 2 or MPE value less than 0.1 represents that the test chemical is non-phototoxic, PIF >2 and < 5 or MPE > 0.1 and < 0.15 predicts that the test chemical is probable phototoxic, and PIF over 5 or MPE > 0.15 represents that test chemical is phototoxic (Guidelines 2019).

6.5 Skin Models for Phototoxicity

Several models have been developed to assess the phototoxicity potential of cosmetic formulation. Earlier, in vivo models were used for phototoxicity testing, but testing was banned due to ethical concerns. In vitro models are currently being used, which can be divided into two based on the number of skin cell types.

6.5.1 2D Skin Models

Cell cultures have been used since the early 1990s by scientists. Cell culture refers to cells obtained from living tissues grown under a controlled environment. If grown in flat climates such as petri dish are called 2D cell cultures. Initially, keratinocytes present in the epidermis layer of the skin were used as the primary cell type for the cell culture. The growth of cells outside the organism's body requires specific nutrients and equipment to survive and divide. Cells are provided with complete media that contains amino acids, buffers, vitamins, and antibiotics (Breslin and

O'Driscoll 2013). Various types of 2D skin models have been developed, which use specific cell types to study the toxicity of chemicals in our environment.

6.5.1.1 NIH 3 T3 Cell Line

Primary fibroblast cell line recommended by OECD for in vitro phototoxic studies, established by scientists George Todaro and Howard Green in 1963, is derived from mouse embryonic fibroblasts. NIH 3 T3 cells are named after their "3-day transfer, inoculum 3 × 105 cells" culturing protocol. Cells are cultured in flat cell culture flask coated with poly-L lysine or collagen for adhesion (Littlefield 1982). Phototoxicity testing is based on the cytotoxicity of a chemical in the presence and absence of a non-cytotoxic dose of UVR/visible light. Cytotoxicity is expressed by dependent concentration reduction in uptake of neutral red dye. Cells are first incubated with test chemicals for an hour, and a half of the plates are exposed to a non-cytotoxic dose of UVR/visible light, and cells are incubated for 18–24 h. NRU determines cell viability. The NIH 3 T3 cell line is also used for various cosmetic ingredients' phototoxicity testing.

6.5.1.2 HaCaT Cells

HaCaT cell line is immortalized human keratinocytes cells and keratinocytes representing the significant cell type present in the epidermis, used as a screening tool for predicting the phototoxicity and irritation potential of surfactants, cosmetic ingredients, drugs, and herbal formulations. MTT and NRU assays are performed to test any phototoxic compound. HaCaT provides reproducible and reliable results (Shukla et al. 2022).

6.5.2 3D Skin Models

To overcome the limitations of 2D skin models for phototoxicity, researchers are investigating the application of a 3D reconstructed human epidermis model. The phototoxicity potential of any chemical is evaluated by comparing the viability of reconstructed human epidermis tissue (RHE tissue) with test chemicals in the presence and absence of UV/Vis radiation or sunlight. The significant advantage of 3D RHE tissue is that the test chemical is applied topically to the tissue (Tavares et al. 2020).

Hydrogel systems are the most dominant technique for creating 3D cultures. It serves as a scaffold for dermal fibroblast co-cultured with keratinocytes on the top. Generally, the hydrogel material is collagen I, but other extracellular matrix proteins can also be used. The hydrogel system allows the differentiation of keratinocytes into different layers using high Ca^{2+}, low temperature, and a unique protocol for air exposure. Cell types are generally primary keratinocytes and primary human dermal fibroblast, but HaCaT can also be used. Hydrogel models can be used to study the phototoxicity of cosmetics and other chemicals (Stanton et al. 2015).

3D skin models can be produced by 3D bioprinting; they can create different layers of the desired material in the form of hydrogel or biodegradable scaffold. Cells

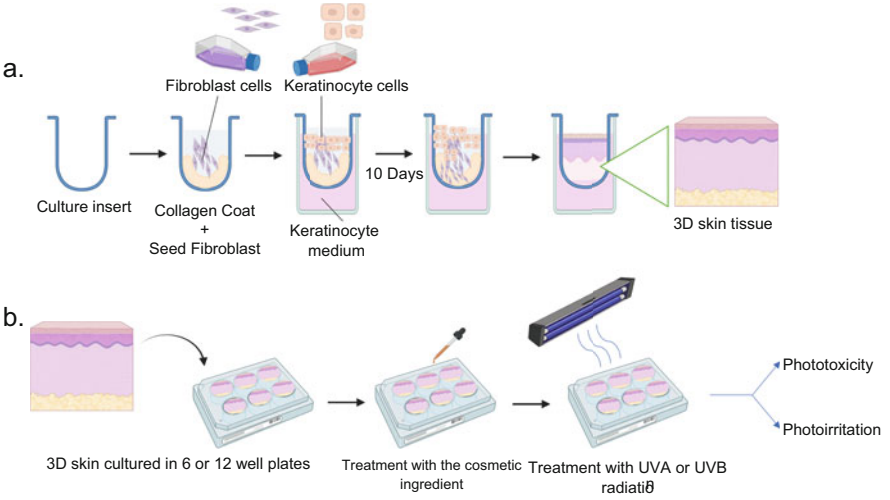

Fig. 6.3 3D skin model. (**a**) Development of 3D skin model. (**b**) 3D skin model used for phototoxicity assessment

or biomolecules can later be added to the scaffold to form biological structures (Murphy and Atala 2014). Bio-printed skin was generated, having complex systems with epidermal and dermal layers containing keratinocytes, fibroblasts, melanocytes, and collagen. 3D bioprinting is automated and has high reproducibility and throughput, but the cost of production is increased. 3D bio-print can be used to assess the cytotoxicity potential of cosmetic formulations and further for the phototoxic potential of cosmetics. Another method for 3D cell culture is a microfabricated system or cell on a chip that provides the required nutrients using microfluidics. It comprises a microfabricated cell culture of keratinocytes and fibroblasts with microfluidic channels. Three major models are currently used in phototoxicity testing (Fig. 6.3).

6.5.2.1 SkinEthic 3D Model

The SkinEthic model has a high similarity to the human epidermis. Layers of the epidermis, such as stratum corneum, stratum granulosum, and stratum spinosum, can be found in the 3D tissue. The model can be defined as "epidermis reconstituted by air lifted culture of normal human keratinocytes for 17 days in chemically inert polycarbonate filters." The number of layers of stratum corneum differs from the native skin issue. Major ceramides and their precursor, glucosylceramides, are present in the SkinEthic model. The general composition of lipids in the SkinEthic model is similar to native skin. In native skin tissue, the stratum corneum is maintained at constant thickness due to desquamation, but in the culture model, the stratum corneum becomes progressively thicker (Pellevoisin et al. 2018).

Phototoxicity testing was assessed by comparing the results with in vivo data. Several phototoxic and non-phototoxic compounds were tested with the model, and it could discriminate between phototoxic and non-phototoxic compounds. Leakage

of LDH was used as a marker for the decrease in cell viability, and increased release of IL-8 and expression of IL-8 mRNA was used to quantify the phototoxicity of test compounds. It is shown that the SkinEthic model was working correctly.

6.5.2.2 EpiSkin 3D Model

EpiSkin model can be defined as a "type I bovine collagen matrix, representing the dermis, surfaced with a film of type IV human collagen, upon which is laid, after 13 days in culture, stratified differentiated epidermis derived from second passage human keratinocytes." EpiSkin has two models: the epic skin irritation model and the EpiSkin penetration model. The stratum corneum of the EpiSkin model has more layers than native skin. The organization of cells in the epidermis layer differs slightly from the native epidermis. The shape of cells in different layers is somewhat different. Lipid composition is close to that of the human epidermis. Phospholipids were comparatively less in the penetration model but almost the same in the irritation model. Precursors of ceramides and glycosphingolipids were also comparable to the human epidermis.

To examine the model for its phototoxicity testing, several known phototoxic compounds such as chlorpromazine, ofloxacin, and 6-methyl coumarin were used as controls. They were exposed to UVA at a non-cytotoxic dose for 1 h. MTT cell viability test was performed after incubation, and IL-1α release into the culture medium was quantified. Increased cell mortality and a rise in IL-1α release confirmed the model's ability to identify phototoxic compounds. Irritation and corrosivity testing was performed, and the results demonstrated that the model could distinguish between irritants/corrosive and non-irritant/non-corrosive chemicals (Lelièvre et al. 2007).

6.5.2.3 EpiDerm

EpiDerm was created by MatTek corporation and introduced in the market in 1993. The model can be described as "normal, human-derived epidermal keratinocytes (NHEK) which have been cultured to form a multilayered, highly differentiated model of the human epidermis." General morphology is similar to that of native epidermis tissue. The only difference is that the rete ridges present in the native tissue anchor the epidermis and dermis, which are absent due to being grown on polycarbonate filters. The cell shape is columnar to round and flattened in the stratum spinosum. Lipid content is similar to that of the regular human epidermis.

Phototoxicity testing of the EpiDerm skin model was done by topically applying test materials with five different concentrations and then exposed to a non-cytotoxic dose of UVA. In visible light, 1 day after the irradiation, cytotoxicity was determined by MTT assay, and the model was successfully able to differentiate between phototoxic and non-phototoxic compounds. Irritation testing was done using a formulation containing surfactants, and IL-1α mRNA levels determined the level of irritation. Results differ from human skin. The barrier function of the model is suboptimal compared to human skin. Therefore, the model can be used for screening possible irritants. Comparison studies of 22 cosmetic formulations for irritancy were done in vivo human skin and the EpiDerm model. The results indicated that the

model could be used to assess the irritancy potential of cosmetic formulations. 3D skin models provide a more accurate representation of the environment of human skin (Cannon et al. 1994).

6.5.3 In Vitro 2D Vs 3D Models

2D cell culture differs from 3D cell culture with respect to cell morphology and other features. Primarily, 2D culture is grown in flat environments such as Petri plates, while the 3D cell culture is shaped into 3D spheroids using specialized conditions for culturing.

The significant difference between the two is that 2D cell culture does not represent the environment of in vivo processes. The single-cell type culture, such as fibroblast or keratinocytes, lacks the interaction, such as cell-to-cell or cell-to-extracellular matrix. At the same time, 3D cell culture gives a good representation of in vivo processes and provides a better understanding of cell-to-cell or cell-to-ECM interactions. 3D models also have gene expression capabilities, thus mimicking the in vivo process (Teimouri and Agu 2016).

The presence of stratum corneum in 3D models allows the test of cosmetics products or drugs to study skin barrier penetration. This also allows for the application of cosmetics directly onto the top layer (stratum corneum). At the same time, the 2D model lacks the barrier function, and only soluble compounds in an appropriate solvent can be used.

2D cell cultures are cheap, have ease of use, and a vast resource of scientific literature makes them more advantageous over 3D. 3D cell culture is more costly, harder to maintain, and not widely used worldwide. Another problem associated with 3D cell culture is the inability to remove cellular waste that can get accumulated in the culture. This problem can be solved by using a microfabricated cell culture that provides micro channels that enable the flow of nutrients and remove cellular waste (Kapałczyńska et al. 2018).

6.6 Conclusion

Solar radiation contains UV, visible light, and infrared radiation that interact with the skin. The cosmetic formulation consists of numerous ingredients that may be toxic to the skin. Some of the components may be photolabile. When these chemicals are exposed to sunlight, they become phototoxic. It is essential to check the safety of the chemicals in cosmetic formulations. Several models are being used to evaluate the phototoxicity potential of cosmetic ingredients. 2D models have been used for this purpose for several years. It is cheap, easy to maintain, and provides reliable results, but the major problem is that it does not represent the actual human skin. The skin has several layers, each having a specific function. To overcome the problem, 3D skin models are developed that mimic the human skin. Using a 3D skin model, we can also study the absorption and penetration of chemicals in the skin layers and how

it affects the physiology of the skin. 3D models having several advantages are less popular than 2D models because of high maintenance and cost. Each skin model has its advantages and disadvantages. It depends on the study which model is best suited for it.

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