INTRODUCTION
Phytodermatoses are diseases caused by the contact of human beings with plants, whether in professional or leisure activities, and even in their therapeutic use\cite{1-4}.

Many different types of plant dermatitis have been recognized: mechanic by direct contact with the cactus thorns; pharmacological caused by active substances, such as the classic nettle; primary irritant, such as the euphorbia, with its irritant resin; allergic phytophotodermatitis due to type IV hypersensitivity reaction (mediated by cells), a common example is the resin of the Pistacia lentiscus (Mastic); and finally the phytophotodermatitis, the main purpose of this review, which occurs by contact with plants followed by an exposure to sunlight, producing erythema, blisters or hyperpigmentation. This disorder is defined as a phototoxic reaction due to direct effect of light and the photoactive substances, with no immune involvement.

PHYTOPHOTODERMATITIS
Phytophotodermatitis (PPD) was described for the first time by Klaber who introduced the term ‘phytophotodermatitis’ in 1942 and identified the natural psoralens in plants and also isolated beracapten (5-methoxypsoralens) of the bergamot essential oil. This study was the first indication that psoralens are photoactive agents\cite{5}.

Psoralens, also known as furocoumarins, are naturally occurring or synthetic tricyclic aromatic compounds, deriving from the condensation of a coumarin nucleus with a furan ring\cite{6}. They are present in many plants of different families, such as: Umbelliferae, Rutaceae, Moraceae and Leguminosae, mainly Tahiti lemons (Figure...
1), figs and celery. These natural psoralens have been identified as phytoalexins and are important components of plant defense against fungi and insects (6).

PPD is a phototoxic reaction entirely independent from the immune system. This reaction occurs when the skin is exposed to photosensitizer substances and to ultraviolet radiation, different from the photoallergic reactions, in which there is an immunologic component (3,4,7,8).

When a photon with appropriate wavelength joins a psoralen, it is absorbed, releasing energy, it is not clear if in the form of heat, fluorescence (ability of one compound to react to ultraviolet rays) or phosphorescence (ability of a chemical species to emit light), forming what is called a photo-product.

PPD has being described in many countries such as the United States, Canada, Germany, United Kingdom, Italy, Holland, Dinamarca, Belgium, Spain, Greece, Turkey, Israel, Singapore and Korea.

Most of these publications focus on the clinical diagnosis, with the hands being the most common site due to the manipulation of plants (Figure 2). In South Brazil the Tahitian lemons are the most common cause of photocotoxic reaction. PPD has a wide range of clinical presentations (9-23) may appear as a “bizarre burns” (Figure 3a) that are sometimes mistaken for child abuse (9) or for lymphangitis (19), due to its red streaks (Figure 3b).

Severe burns caused by the ficus leaves used as ‘suntan lotion’ were also reported (24,25), a patient exhibited complications such as hemolytic anemia and retinal hemorrhages. There is one report describing PPD in animals (26).

Clinical publications are rare, probably due to the easy diagnosis. There are some review studies (3,4), in which the most common causes attributed to PPD are the citric fruits such as tangerine (Citrus bergania), lime (Citrus limetta), Tahitian lemon (Citrus medica), lemon (Citrus limmonia). Other causes include celery (Apiumgraveolens), carrots (Daucuscarota), garden rue (Rutagraceolens), figs (Ficuscarica), true cinnamom (Cinnamomumzeylanicum), parsley (Petroselinum sativum), breadnut (Brosimumgaudichaudii), cumaru (Amburanacearensis), mountain arnica (Arnica montana), garden angelica (Angélicaoficinallis) (4) and mango tree (3).

Similar to some reports from other countries, there is variable etiology, depending on the flora and habits of each region. In South Brazil, the most frequent cause is the Tahiti lemon. During summer months, its occurrence is higher because the sunlight is more intense and there are more outdoor activities.

It is possible to find several clinical patterns of PPD (77), such as linear vesicular lesions known as meadow grass dermatitis (Dermatitis bulbosa striata pratensis); lesions in the cervical region by using perfumes with citrus essences; the berlock dermatitis; and the typical PPD that occurs on the back of the hands due to the manipulation of lemons. Acute erythema with blisters (Figure 2) or only a less severe picture with hyperpigmentation can be found on the hands. Sometimes psoralens are applied or splashed in other areas, leading to lesions with atypical configuration (Figure 3A).

Post-inflammatory pigmentation occurs by two mechanisms: (1) pigment incontinence secondary to epidermal necrosis and (2) increase in the number of functional melanocytes and melanosomes, similar to what occurs in PUVA therapy (28).

There are very few information on the histological findings of phototoxic reactions, maybe because the diagnosis is established clinically (28-31). Epithelial degeneration was reported in a publication using light microscopy (20).

![Figure 1 Cross-section of Tahitian lemon, note the release droplets upon its manipulation (arrow), which contain phototoxic substances.](image1)

![Figure 2 Acute phase. Erythema and blisters (arrow) on the hands in the severe phase of PPD, more intense on the right hand.](image2)

![Figure 3 A. Bizarre hyperchromic lesions in the resolution phase on the abdomen, touched by the hands after handling lemons. B. linear lesion simulating lymphangitis.](image3)

![Figure 4 Erythematous areas on the epilated dorsum of the rat 48 hours after being sprayed with Tahiti lemon peel juice.](image4)
EXPERIMENTAL STUDIES ON PPD REPRODUCTION

PPD was experimentally reproduced in some studies: the first was a Brazilian experimental study on humans in the 70s performed with artificial light. In this study only the peel juice of the Tahitian lemon triggered PPD. Biopsies were not performed[32].

In the same decade, another study produced PPD using crushed leaves of Dictamnus albus and reproduced PPD lesions applying ultra-violet in the subjects. An occlusion time was 30-120 min and even after freezing of the plant material was able to evoke PPD. No histological analyses were performed[33].

Another study on humans was published in 1983 with Heracleum laciniatum. The best long-wave ultraviolet light that produced PPD was identified in the range 315-375 nm, with peak sensitivity at 330-335 nm, therefore, within the ultraviolet A spectrum[34]. Subsequently, the same authors identified the flowers and leaves as the best parts to trigger PPD[35].

Several animal models have been used to investigate the phototoxic reactions to medications[36-40].

PPD was experimentally reproduced in rats using sunlight with Tahiti lemon peel juice[41] (Figure 1). Very short exposure time (2.5 minutes) was enough to induce it. The light microscopy revealed an epithelial lesion that appeared in 24 hours, however, the erythema was only clinically evident on the dorsum of the animals after 48 hours (Figure 4).

In another study, with exposure times ranging from 5 to 8 min, a serial histological study on the epidermal changes immediately after induction was performed: after 1, 2, 4, 6, 24, 48 and 72 h, the left half of each rat was used as the control and was only exposed to sunlight; in another area only peel lemon juice was applied[42].

Similarly to the first study, no lesions were detected using light microscopy before 24 hours in areas where PPD was reproduced. At 24 hours, there were no changes in controls exposed to sunlight (Figure 5A) and in the induced PPD keratinocyte necrosis and epidermal vacuolization were visible (Figure 5B). At 48 hours, no significant changes were observed in the controls; there was significant epidermal vacuolization with intra and subepidermal cleavages in induced PPD (Figure 6). These changes were less intense in 72 hours. No clinical or histological lesions were seen on the control side of the rats.

In a third evaluation with an animal model, transmission electron microscopy was used to verify whether this more sensitive method could detect histological changes immediately after the induction, and after 1 to 2 hours i.e. before lesions were detected by a conventional light microscopy[43].

Vacuolization and membrane ruptures were identified (Figure 7). As in light microscopy there were no changes in the controls (Figure 8A). There were desmosomal lesions characterized by isolated desmosomes, no longer attached to the keratin filaments (Figure 8B) in the experimental PPD. At higher magnification, cell membrane ruptures and free desmosomes could be seen (Figure 9).

DISCUSSIONS

Animal models are used in experimental studies on phototoxic dermatoses[36-40], including PPD[41-43], which could be successfully reproduced in rats.

PPD was reproduced in the experimental model using only the lemon peel juice, in accordance the results obtained with the experimental studies on humans[32] and consistent with the findings of Figure 8 Light Microscopy. A: Control area exposed only to sunlight after 24 hours: epidermis with no significant changes. B: Induced PPD after 24 hours: keratinocyte necrosis (arrow) and vacuolization (HE 400x).

Figure 6 Light Microscopy. Induced PPD after 48 hours. A: Confluent cytoplasmatic vacuolization and epidermal necrosis. B: subepithelial blister formation (HE 400x).
Figure 7 Transmission electron microscopy immediately after the experimental induction of PPD. A: Basal keratinocyte with vacuolization (arrows) (×6,000). B: Detail showing cell membrane rupture (arrow) (× 40,000).

Figure 8 Transmission electron microscopy. Normal desmosomes in control-rats with insertion of keratin filaments (KF) in the desmosomal plaques (arrows) (× 43,400). Vacuolization of the intercellular spaces with disperse desmosomes (arrows), with loss of keratin filament insertion (KF), 1 hour after the experimental induction (× 35,000).

Figure 9 Transmission electron microscopy 2 hours after the PPD experimental induction. Detail of desmosomal lesion without contact with keratin filaments and discontinuity of cell membrane (arrow) (× 47,800).

Figure 10 Schematic chronological evolution of phytophotodermatitis.

In another study, in which the psoralens in the lemon peel were found in concentrations 13 to 182 higher than in the fruit juice[44].

In the first hours after the experimental induction of PPD, the histological and clinical evaluation showed the epidermis to be normal. Epidermal necrosis was detected at 24 hours and they progressed to intra and subepidermal blistering in 48 hours. Although the histological aspect showed to be normal before 24 hours, lesions became visible some hours later. This is in agreement with knowledge on cell death and apoptosis. It is known that there is an interval between the harmful stimulus and the morphological manifestation of the injured or dead cell, also seen with some drug reactions. In vitro cell culture models used in experimental studies have also induced cell apoptosis with exposure to psoralens and ultraviolet A[6].

With transmission electron microscopy, which can detect early changes, it was possible to immediately identify peripheral
keratinocyte vacuolization and membrane lesions.

One to two hours after the experimental procedure, lesions become quite evident showing membrane ruptures and desmosomal degeneration. Desmosomes were seen to be rounded with the cell membrane folded over the plaques. These changes progressed to apoptosis and blisters. Figure 10 shows the chronological evolution of microscopic and clinical findings in experimental PPD. Although the use of animal models show limitations, it is possible to demonstrate lesions to the membrane, cytoskeleton, desmosomes, which lead to cell death.

These informations not only account for part of the PPD pathogenic phenomena, but could be used in understanding the use of photodynamic therapy\(^{36,60}\), in which lesions of epithelial origin are destroyed, considering the principle of interaction between photoactive substances and light sources.

**CONFLICT OF INTERESTS**

The authors declare that they do not have conflict of interests.

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