Skin Biopsy

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Abbreviations

AP	Anatomopathological examination
DIF	Direct immunofluorescence
PNF	Proximal nail fold

Key Points

- Skin biopsy can be defined as a surgical procedure to remove a skin fragment for the purpose of laboratory analysis.
- The most commonly used exam to evaluate biopsy skin material is histopathology.
- Most skin biopsies are performed using local infiltration anesthesia.
- There are many types of skin biopsy, punch, curettage, shave, saucerization, and elliptical biopsy.
- The technique used to perform nail biopsy depends on the location of the lesion and its clinical aspect.

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Introduction

Skin diseases are very prevalent and often difficult to differentiate with clinical examination only, making skin biopsy an important aid in dermatology practice.

Skin biopsy can be defined as a surgical procedure to remove a skin fragment for laboratory analysis. The biopsied fragment may represent part of the lesion (incisional biopsy) or the entire lesion (excisional biopsy).

This procedure is usually followed by a histopathological examination to clarify and/or confirm a diagnosis, classification and/or staging of certain diseases, evaluation of surgical margins, and treatment of skin neoplasms.

Choice of Lesion Area for Biopsy

The biopsy should ideally be undertaken on a lesion which is most typical and representative of the underlying pathology. The ideal fragment should preferably include an elementary lesion with no evidence of secondary lesions, such as excoriation, secondary, or partially treated infection (previous use of topical medication). Whenever possible, all skin layers (epidermis, dermis, and hypodermis) should be included. In certain cases, it is important to include perilesional skin for histological comparison (Table 79.1).

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Table 79.1 Type of lesion and appropriate area for diagnosis

Type of lesion	Biopsy-appropriate area
Bullous	Full blister or border for anatomopathological examination (AP) Perilesional area for direct immunofluorescence (DIF)
Vasculitis	More than 1 lesion, with different times of duration, preferably choose: Lesion with >72 h for AP Lesion with <24 h for DIF
Panniculitis	Deep biopsy containing subcutaneous tissue
Polymorphous, circinate	More "active" peripheral border
Ulcer	Ulcer edge associated with surrounding skin
Pigmentation disorders (e.g., vitiligo)	Two fragments—1 fragment of normal skin and 1 fragment of the most hypo/achromic area
Pigmented lesions (suspected melanoma)	Excisional biopsy
Tumoral	Thickest area, avoid necrotic area
Cicatricial alopecia	Active lesion area (erythema, perifollicular desquamation)
Non-cicatricial alopecia	Injury already established When androgenic alopecia is suspected, two fragments—one of a typical area and another of an unaffected area (e.g., occiptal)

In case of dermatosis with lesions in different areas, the choice of the body region to be biopsied is also important as, for instance, lesions extracted from areas below the waist are more difficult to be interpreted because they are subject to alterations caused by blood stasis [1]. Other unfavorable areas for analysis of microscopic changes are body regions subject to constant friction, such as knees and elbows, as the skin becomes thick in these areas, and regions under intense sun exposure, due to elastosis [2]. The face and neck should also be avoided for esthetic reasons, as well as areas on joints and legs, which are harder to heal.

Complementary Research

Below we list the main complementary exams performed after a skin biopsy. Bearing in mind that it is extremely important to provide the pathologist with as much information as possible, including the patient's age, sex and Fitzpatrick´s phototype, a summary of the clinical picture, biopsy location and technique, and diagnostic hypotheses.

- **Histopathology:** This is the most commonly used exam to evaluate biopsy material. Useful in the diagnosis of most pathologies, the most widely used stain being hematoxylin-eosin. The solution for specimen preservation until the exam is processed is 10% neutral, preferably buffered, formalin.
- **Direct immunofluorescence:** Useful in the diagnosis of autoimmune bullous diseases, vasculitis, and genodermatoses. The solution for specimen preservation until the exam is processed is Michel's medium; however, the saline solution may be used when processing begins before the first 48–72 h.
- **Culture:** Useful in the diagnosis of infectious diseases caused by fungi, bacteria, and mycobacteria. The solution for specimen preservation until the exam is processed is saline.

Standard Protocol for Skin Biopsies

Free and Informed Consent: After the patient is informed about the proposed procedure, it is suggested that they sign a term of free and informed consent, specifying the technique to be used, potential risks and complications inherent to the procedure, as well as their consent and understanding of all risks.

Photography: A photograph is taken of the area to be biopsied and the lesion to be removed, always upon the patient's prior written authoriza-

tion. The authorization for photography can be included in the term of consent.

Marking: Prior marking of the site to be biopsied with a surgical pen, as some lesions, especially the most vascularized ones, may become less evident or even disappear momentarily after anesthesia.

Asepsis: Asepsis can be performed with 2% chlorhexidine in alcoholic or aqueous solution, povidone-iodine or 70% isopropyl alcohol. The use of chlorhexidine was shown to be superior to other antiseptic agents, considering antimicrobial spectrum and duration. We must pay special attention when using chlorhexidine in regions close to the eyes and ears, due to the risk of keratitis and otitis [3].

Anesthesia: Most skin biopsies are performed using local infiltration anesthesia [4]. The anesthetic solution must be injected into the dermis (more superficial lesions) or subcutaneously. The most commonly used anesthetic is 0.5–2% lidocaine, which may be associated with epinephrine 1:100,000–1:200,000 to reduce bleeding and increase the duration of anesthesia. To reduce anesthetic pain, due to the acidic pH of the lidocaine-epinephrine solution, 8.4% sodium bicarbonate can be added (solution: 1 part of sodium bicarbonate to 10 parts of lidocaine-epinephrine), which increases the pH to a more physiological level, and significantly reduces anesthetic pain [5, 6]. The use of syringes smaller than 1–3 mL and thinner needles (30G), as well as a slow injection speed, also reduce the pain of infiltration [4].

Types of Biopsy

After applying the standard protocol (term of consent, photograph, marking with a surgical pen, asepsis, and anesthesia), we proceed with the biopsy procedure of choice.

Punch

Punch is a pen-shaped surgical instrument which contains a cylindrical or conical blade at one end, with diameters ranging from 1mm to 10mm. For most biopsies, a 3–4mm punch is sufficient, but eventually punches with larger diameters can be used when a greater tissue representation is necessary. They can be disposable or reusable (stainless steel) (Fig. 79.1).

A punch is preferably used to perform incisional biopsies, but in smaller lesions it can be used for complete removal.

Technique

To perform a punch biopsy, place the blade in contact with the skin at a 90° angle, and then rotate the instrument on its axis, always in the

Fig. 79.1 Punch reusable n.2 and punch disposable n.4

same direction and applying pressure onto the skin, so that it can cut through the skin until it reaches the desired depth.

Carefully remove the material with tweezers, cut the base with scissors or with the punch blade at a 45° angle [7].

Immediately after removing the specimen, place it in the appropriate conservation medium for subsequent analysis. It is usually recommended to suture the surgical wound with one or two simple stitches, but secondary intention healing can also be used (Fig. 79.2).

Curettage

Curettage consists of scraping lesions with a curette, a surgical instrument whose sharp end has a circular or oval blade. For most dermatological procedures, curettes of 2–5mm are used. Curettes can be disposable or reusable (stainless steel).

This is preferably used to treat epidermal lesions that have some type of cleavage plane with the dermis (e.g., molluscum contagiosum, seborrheic keratoses, common warts, among others)

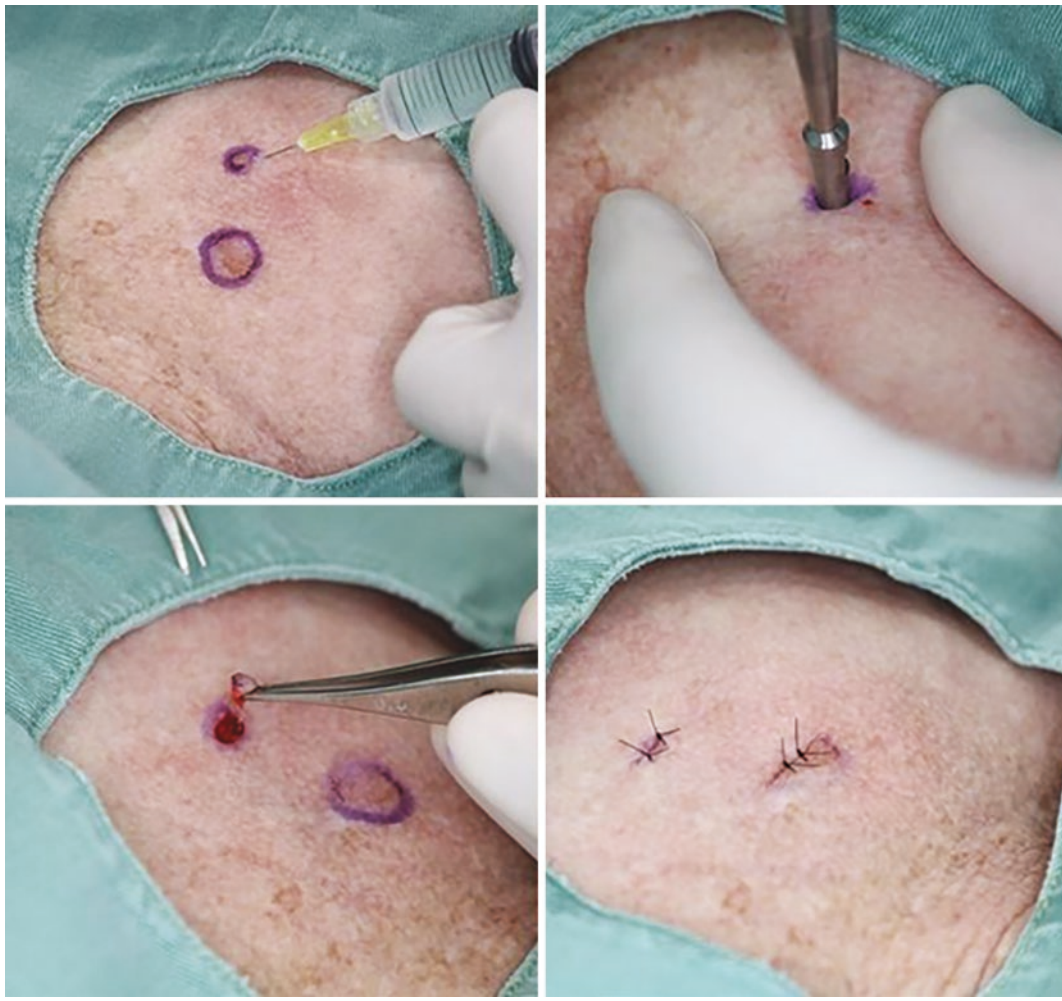


Fig. 79.2 Punch technique

[8–11]. It can also be an option for diagnosis and treatment of superficial skin neoplasms, such as actinic keratoses, superficial basal cell carcinomas, and *in situ* squamous cell carcinomas in low-risk areas [12, 13]. However, it should be noted that the technique generally produces fragmented samples, and it is not possible to assess the depth and surgical margins of the lesions [8, 9].

Technique

The skin around the lesion should be stretched with the non-dominant hand and the lesion should be scraped with the curette, with movements parallel to the skin and applying light to moderate pressure, until complete removal of the lesion.

Hemostasis can be performed using compression dressings, electrocoagulation, or chemical cautery (e.g., aluminum chloride 25–40%).

Healing will occur by secondary intention, generally with good cosmetic results, and mild dyschromia (tendency to hypochromia) may be observed.

The technique is not usually applied in areas of lax skin or that tear easily (e.g., eyelids, elderly patients' hand dorsum, among others) [8] (Fig. 79.3).

Shave Biopsy

This consists of the tangential excision of skin lesions. It can be performed with scalpel blades, flexed razor blades, or scissors, for diagnostic or therapeutic purposes.

Shavings are usually reserved for the removal of benign exophytic lesions (e.g., intradermal or compound nevi, acrochordons, fibrous papules of the nose, seborrheic keratoses) [8–11].

Technique

An anesthetic injection is applied beneath the lesion to slightly elevate its base in relation to the surrounding tissue. After choosing the surgical instrument (blade or scissors), the lesion is incised tangentially to the skin (Figs. 79.4 and 79.5).

Hemostasis can be performed using compression dressings, electrocoagulation, or chemical cautery (e.g., aluminum chloride 25–40%). It is a fast and effective method, which usually does not require suturing.

Healing will occur by secondary intention, usually with good cosmetic results, and mild hypochromia may be observed. Depressed scars can result when we raise the base of the lesion more than necessary when creating the anesthetic button.

In lesions with a minimally narrow pedicle (e.g., acrochordons), the procedure can be performed without anesthesia, with minimal pain.

Deeper shavings are performed with greater blade angles and are known as *saucerization*.

Fig. 79.3 Curretage of a viral wart

Deep Shave Biopsy/Saucerization

Deep Shave biopsy can be an adequate technique for flat lesions without evidence of vertical growth. In addition to being quick to perform and providing good esthetic results, it presents an adequate tissue sample, diagnostic accuracy, and the possibility of safety assessment of the deep margins of flat lesions [14, 15].

The ideal biopsy technique should be easy and quick to perform (to facilitate generalized application), be associated with minimal morbidity, allow for precise staging of lesions considered to be malignant, and not compromise long-term oncological results [16].

Technique

Infiltration of local anesthetic is performed, application of a thin layer of ointment on the lesion to firm the tissue, followed by the removal of a disc of tissue using a scalpel or curved razor blade (Fig. 79.6). It produces a sample extending to the upper dermis or the mid-dermis, depending on the angle of the blade. Hemostasis is performed with the application of aluminum chloride and a compression dressing for 24 h. Placing the material on a firm surface (e.g., suture strand

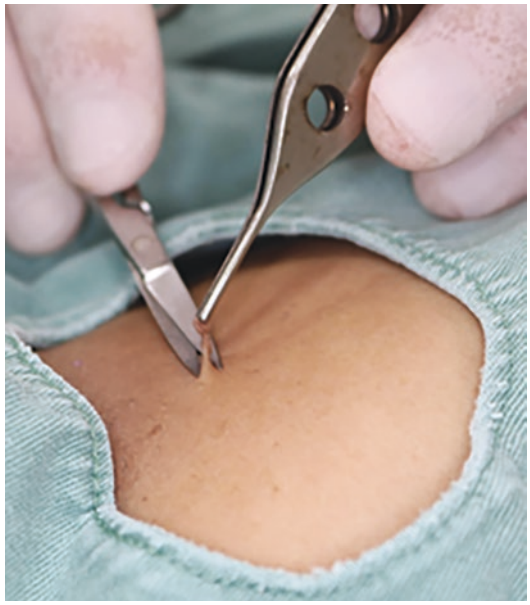


Fig. 79.4 Shave biopsy with scissor

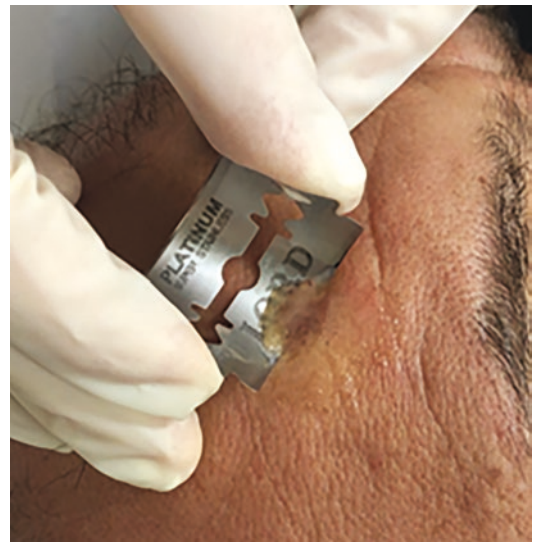
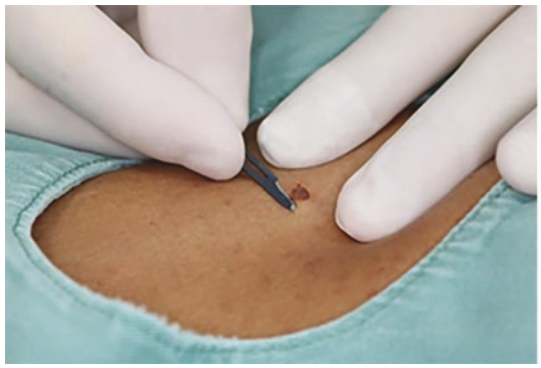


Fig. 79.6 Deep shave biopsy



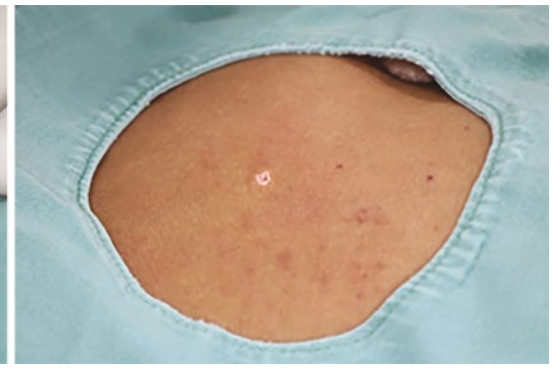


Fig. 79.5 Shave biopsy with blade n.15

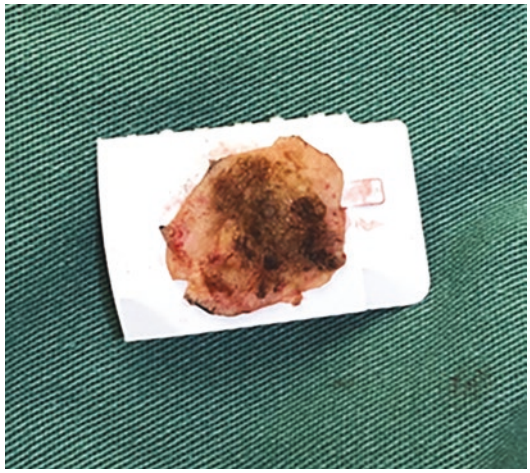


Fig. 79.7 Material on a firm surface

paper) allows keeping it stretched, without curling thus facilitating histological preparation (Fig. 79.7).

Elliptical Biopsy

This consists of the removal of part (incisional biopsy) or all (excisional biopsy) of the cutaneous lesion, in the shape of an ellipse, using a scalpel blade. This technique has the advantage of removing all skin layers.

In cases with clinical suspicion of melanoma, excisional biopsy is considered the gold standard for the possibility of extensively assessing the removed tissue and accurately defining cell atypia and the presence or absence of basement membrane invasion, Breslow index, and other prognostic factors involved in melanocytic neoplasms [14]. However, there are some cases in which it can cause significant esthetic and functional damage, for example, in large, unresectable pigmented lesions and in areas that are difficult to approach, such as the face. In these cases, incisional biopsy guided by dermoscopy has been the recommended initial approach [17]. Partial biopsies are considered safe, not affecting prognosis [15, 18, 19].

Technique

Make the incision as a single continuous sweep, down to fat, rather than a series of small nicks, and hold the blade at 90° to the skin, not angled inwards, so that the ellipse sides will be vertical [20, 21]. The ellipse length should be approximately three times its width to produce an ellipse angle of approximately 30° and prevent buckling when the wound is sutured [21, 22]. The fat under the ellipse should be cut through using scissors, while the ellipse is gently pulled away from the skin using proper forceps. Close the wound using both subcutaneous and surface sutures, if necessary, employing the appropriate suture technique [21] (Fig. 79.8).

Nail Biopsy

The technique used to perform nail biopsy depends on the location of the lesion and its clinical aspect.

It can be performed with a punch (with or without previous nail avulsion), scalpel (elliptical or tangential excision in the nail bed or matrix), or curette. In the nail bed, excision should generally be along the longitudinal axis, respecting a 4 mm diameter, with or without previous avulsion of the nail plate. In the nail matrix, the excision must be along the horizontal axis, respecting a 3mm diameter. When biopsy of the matrix is performed, the patient must be previously informed about the possibility of permanent nail dystrophy.

Examples of lesions that require biopsy are melanonychia suspected of melanoma, tumors such as squamous cell carcinoma, acral melanoma, glomus tumor, and pyogenic granuloma, as well as inflammatory dermatoses such as psoriasis and lichen planus [23].

We will describe the technique for nail matrix biopsy.

Technique

After asepsis with alcoholic chlorhexidine solution, proximal or distal regional block is performed with local anesthetic (lidocaine), with or without adrenaline. A tourniquet is used on the proximal phalanx with a penrose drain or glove finger for vascular compression. After preparation of the sterile surgical field, the skin is incised

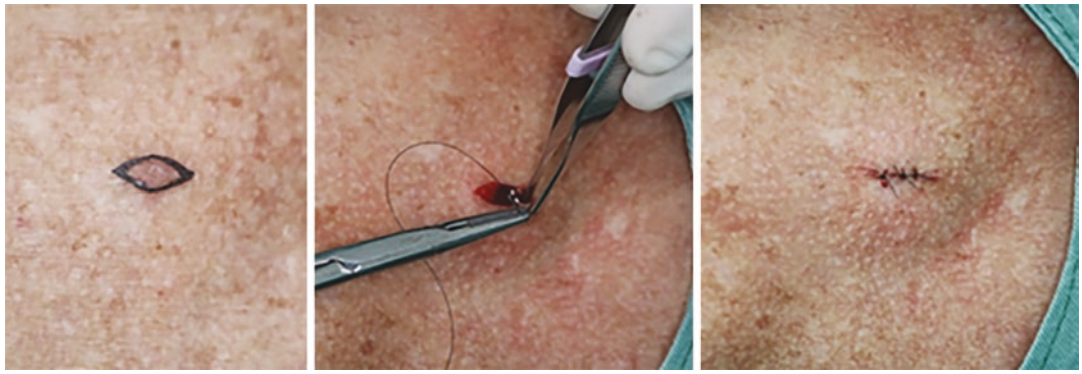


Fig. 79.8 Elliptical biopsy

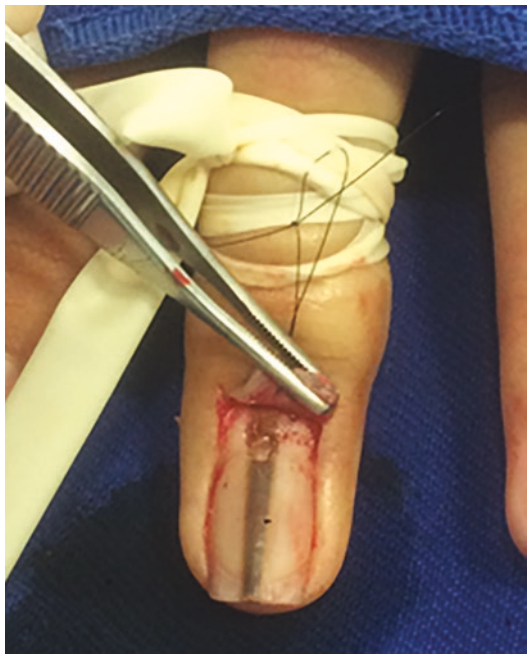


Fig. 79.9 Exploration of the nail matrix, with incisions in the proximal nail fold

with an 11 or 15 scalpel blade for the rebating of the proximal nail fold (PNF) (Fig. 79.9). Then the nail matrix is exposed, and the site is chosen for biopsy with a punch or blade. It may be necessary to remove the nail plate or to perform a transfixion biopsy, without removing it. In small lesions, healing can be by secondary intention or hemostasis with the application of aluminum chloride (35%). For bigger lesions, suture with vicryl 4.0 (biopsy site), suture of the incision edges with mononylon 4.0, followed by removal of the tourniquet, and compression dressing for 24 h [23].

Despite many controversies about the risk of using vasoconstrictors associated with local anesthetics, there is evidence that the association of lidocaine and adrenaline in the usual dosages can be safely applied in patients without peripheral vascular disease [24].

Glossary

Breslow index The measurement of the depth of the melanoma from the surface of your skin down through to the deepest point of the tumor.

Curette A surgical instrument whose sharp end has a circular or oval blade

Histopathology Exam to evaluate biopsy material under a microscope, useful in the diagnosis of most pathologies, the most widely used stain being hematoxylin-eosin.

Immunofluorescence assays (IF) Are an important tool for diagnosing acquired autoimmune blistering diseases, since they detect "in vivo" autoantibodies. There are two main subtypes: direct immunofluorescence (DIF), which is performed on perilesional skin or mucous membranes to detect tissue-bound autoantibodies; and indirect immunofluorescence (IIF), that quantifies a patient's circulating autoantibodies, utilizing human foreskin or monkey esophagus as substrates.

Melanonychia Brown or black discoluration of a nail. It may be diffuse or take the form of a longitudinal band.

Nail matrix Consists of specialized cells that produce the nail plate. It is located at the end of the digit (finger or toe) under the skin beyond the distal phalangeal joint.

Punch A pen-shaped surgical instrument which contains a cylindrical or conical blade at one end.

Saucerization/deep shave biopsy Deep tangential excision of skin lesions.

Shave biopsy Tangential excision of skin lesions.

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