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REVIEW

The Pathogenic Role of *Demodex* Mites in Rosacea: A Potential Therapeutic Target Already in Erythematotelangiectatic Rosacea?

Fabienne M. N. Forton

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ABSTRACT

Rosacea is a common facial dermatosis but its definition and classification are still unclear, especially in terms of its links with demodicosis. Triggers of rosacea (ultraviolet light, heat, spicy foods, alcohol, stress, microbes) are currently considered to induce a cascading innate and then adaptive immune response that gets out of control. Recent histological and biochemical studies support the concept that this inflammatory response is a continuum, already present from the onset of the disease, even when no clinical signs of inflammation are visible. The *Demodex* mite is beginning to be accepted as one of the triggers of this inflammatory cascade, and its proliferation as a marker of rosacea; moreover, the papulopustules of rosacea can be effectively treated with topical acaricidal agents. *Demodex* proliferation appears to be a continuum process in rosacea, and may not be clinically visible at the onset of the disease. Molecular studies suggest that *Demodex* may induce tolerogenic dendritic cells and collaborate with vascular endothelial growth factor (VEGF) to induce T cell exhaustion and favor its own proliferation. These interactions among VEGF, *Demodex*, and immunity need to be explored further and the nosology of rosacea adapted accordingly. However, treating early rosacea, with only clinically visible vascular symptoms, with an acaricide may decrease early inflammation, limit potential flare-ups following laser treatment, and prevent the ultimate development of the papulopustules of rosacea. The effectiveness of this approach needs to be confirmed by prospective controlled clinical trials with long-term follow-up. Currently, the evidence suggests that patients with only vascular symptoms of rosacea should be carefully examined for the presence of follicular scales as signs of *Demodex* overgrowth or pityriasis follicularum so that these patients, at least, can be treated early with an acaricidal cream.

Keywords: Benzyl benzoate; Demodicosis; Dendritic cell; Immunotolerance; Ivermectin; MGL; Rosacea; Standardized skin surface biopsy; Tn Ag; VEGF

F. M. N. Forton (✉)
Rue Frans Binjé 8, 1030 Brussels, Belgium
e-mail: fabienne@fforton.be

Key Summary Points

Rosacea is an inflammatory continuum, with all characteristics being already present from the onset of the disease, even if not clinically visible.

Demodex proliferation also appears to be a continuum process in rosacea, and high *Demodex* density is beginning to be accepted as an important trigger of the inflammatory cascade and as a marker of rosacea: moreover, papulopustules of rosacea can be treated using acaricides.

Immunological studies are providing new hypotheses according to which *Demodex* may induce tolerogenic dendritic cells and collaborate with VEGF to induce T cell exhaustion favoring its own proliferation. This proliferation may not be clinically visible initially.

The interactions among VEGF, *Demodex*, and immunity need to be explored, and the nosology of rosacea definitions adapted accordingly.

The effectiveness of treating any patient who only has visible vascular symptoms with an acaricidal cream needs to be confirmed in prospective controlled clinical trials with long-term follow-up, but it is already important to detect patients with pityriasis folliculorum among those with only vascular symptoms of rosacea in order to treat at least these patients with an acaricidal cream.

DIGITAL FEATURES

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INTRODUCTION

Rosacea and demodicosis are common conditions in dermatology practice. While demodicosis is clearly the result of infestation by the *Demodex* mite, the etiology of rosacea is unclear. However, there is increasing evidence to suggest that rosacea is an inflammatory continuum and that there is a key role for the *Demodex* mite in this inflammatory process. In this review, we will analyze these concepts further and discuss the possible implications for definitions and diagnosis, and also for treatment.

This article is based on previously conducted studies and does not contain any new studies with human participants or animals performed by the author.

ROSACEA: DEFINITIONS

Rosacea is a common facial dermatosis with a prevalence of up to 10% if all forms are included [1–4]. Pure vascular rosacea is the most common form, about four times more frequent than rosacea with papulopustules [1]. Because the cause of rosacea is still unknown, rosacea is defined by the presence of non-specific clinical signs and symptoms [5, 6]. Successive expert opinion consensus documents have provided definitions and classifications of rosacea but these remain a source of debate [7–11]. In 2002, the National Rosacea Society (NRS) expert committee defined rosacea as a central face distribution of at least one of four primary features (flushing, persistent erythema, papules and pustules, and telangiectasia) and identified four subtypes: erythematotelangiectatic rosacea (ETR), papulopustular rosacea (PPR), phymatous rosacea, and ocular rosacea [12]. Two key clinical features were considered necessary for a diagnosis of the ETR subtype (flushing and persistent centrofacial erythema) [12]. In the 2018 update, the NRS adopted the suggestions of the global ROSacea COnsensus (ROSCO) panel [13], abandoning the subtypes in favor of

phenotypes, and defining rosacea as the presence of at least one of two core features [phymatous changes and persistent centrofacial erythema (Fig. 1a)] OR two of four major features (flushing, telangiectasia, papules/pustules, ocular manifestation) [6]. In the present review, the abbreviation ETR will be used for the phenotype "rosacea with only vascular symptoms" and PPR for "rosacea with papulopustules".

ROSACEA: AN INFLAMMATORY CONTINUUM

Rosacea is currently considered by most authors as a disease of the immune system, an inflammatory process including innate and then adaptive immune responses, which gets out of control resulting in vascular, inflammatory, and hypertrophic symptoms [2, 5, 14–22]. Genetic (46%) and environmental (54%) influences have recently been demonstrated in a study on twins [23], and many associated co-morbidities have been highlighted [24].

Histological and biochemical studies converge to suggest the continuum of this inflammatory process [25–38]. From the early stages of rosacea, all typical characteristics of the disease are present, although not all may be clinically visible [25]. These characteristics include dilation of blood and lymphatic vessels [26], solar elastosis [25, 27], and increased intradermal fibroblasts [28]. T cell infiltrates are also present from the early stages of rosacea, around intradermal vessels [25, 30], pilosebaceous follicles [25, 31, 32], and sebaceous glands [33]. These infiltrates are essentially composed of Th1 and Th17 type T helper cells (95%) [34–37] and T suppressor cells (5%) [35], but also of mastocytes [33], macrophages and plasmocytes [25], with a $CD4^+$ helper/$CD8^+$ cytotoxic ratio of 2.8, 31% $CD4^+CD25^+$ regulatory cells, and 6% plasmacytoid dendritic cells [38]. This infiltrate, often associated with *Demodex* mites [34, 35, 39], invades the follicular wall and forms granulomas, which have been found in all rosacea subtypes [25, 40, 41]. Expression of the genes encoding the cathelicidin peptide LL-37, a key factor in the pathogenesis of rosacea, and other markers of inflammation are already increased in ETR and even more so in PPR [29], while dermal expression of vascular endothelial growth factor (VEGF) is similarly increased in ETR and PPR [26].

In PPR, this inflammatory reaction reflects a loss of the immunotolerant milieu seen in sebaceous gland-rich zones of heathy skin: dendritic cells become activated and T cells are increased in number and altered to inflammatory type [36].

DEMODEX AND DEMODICOSIS

Demodex folliculorum and *Demodex brevis* are spindle-shaped transparent mites that live exclusively, at low densities, in human pilosebaceous follicles [42–46] as part of the normal adult human microbiota [42, 46–54]. Humans are born without *Demodex* mites on the skin [43, 54], and the mites are progressively acquired by direct contact with the skin of other humans [44, 55]. As a commensal, the *Demodex* mite likely controls the immune system of the host, through undefined mechanisms, to ensure its own survival [8, 10, 56–60].

The delicate host/*Demodex* equilibrium may be tipped in favor of mite proliferation by various factors, including immunosuppression [61–88], diabetes [89–92]), vasodilatory-related factors [8, 10, 25, 27, 31, 57, 93–97], and/or sebaceous hyperplasia [8, 10, 51, 98]. Initially, overproliferation of the mite is not clinically visible, giving rise to what could be called subclinical demodicosis, which can be observed in many skin conditions (including apparently healthy skin and any facial dermatosis), but is commonly encountered in ETR [97] (Fig. 1d). When this proliferation continues, the opisthosomes of the mites become visible to the naked eye, appearing as thin, discreet, regularly dispersed, whitish follicular scales at the base of the hair, often associated with diffuse erythema (which is a key feature of rosacea) [57, 99–102]. These clinically visible symptoms constitute the first stage of demodicosis—pityriasis folliculorum [8, 56, 57, 99, 101–103] (Fig. 1c), called by some primary demodicosis [7]. The symptoms are very discreet and, if the dermatologist is not familiar with the condition and trained to

Fig. 1 Erythema of rosacea and pityriasis folliculorum. **a** Erythema of rosacea on white skin, according to the consensus of the National Rosacea Society (NRS): original photograph published by the NRS [6]. **b** However, as shown on a zoom on the right cheek, this photo clearly reveals the presence of follicular scales, suggesting a diagnosis of pityriasis folliculorum. **c** Demodicosis associated with vascular symptoms of rosacea: discreet thin whitish follicular scales at the base of the hair give a frosted appearance and a rough texture, suggesting a diagnosis of pityriasis folliculorum; this was confirmed by the diagnostic test. Each follicular scale corresponds to the most superficial part of numerous *Demodex* mites agglutinated on a single follicle (blue box). **d** Subclinical demodicosis with vascular symptoms of rosacea: the follicular scales were not detected on close clinical examination, even after cleaning the skin with ether and using tangential illumination, leading to the clinical diagnosis of erythematotelangiectatic rosacea. However, this patient had a high *Demodex* density, suggesting a likely diagnosis of subclinical demodicosis. **e**, **f** Pityriasis folliculorum diagnosed as rosacea and treated with intense pulsed light (IPL): this 41-year-old woman complained of sensitive skin and redness of the whole face for 2 years. She consulted a dermatologist and was treated with isotretinoin for 8 months (30 mg/day for 6 months and 40 mg/day for 2 months) and then by IPL flash lamp (which emits simultaneous wavelengths between 530 and 1200 nm), with no resolution of her problems and even some aggravation. The dermatologist then sent the patient to our clinic for our advice. The patient had diffuse redness all over the face (not shown), more pronounced at the follicular orifices, with slight diffuse edema (visible on the lobule of the ear): the skin appeared irritated. On close examination, there was no vellus hair or follicular scales on the skin of the central face. After the skin was cleaned with ether, two standardized skin surface biopsies were consecutively performed on the right cheek and confirmed the absence of *Demodex* mite ($0 + 0$ D/cm^2). Nevertheless, on small areas not treated by IPL, i.e., the lobule of the ear and the preauricular zone of the cheek, we discovered follicular scales suggesting *Demodex* mites. **e** On the preauricular zone, the mite density was very high, confirming the diagnosis of pityriasis folliculorum. The patient was instructed to apply an acaricidal cream (benzyl benzoate 12% and crotamiton 10% in Cetomacrogol cream) all over the face (not on the eyelashes or the lips) once daily for 1 week, then twice daily. **f** Two months later, facial signs and symptoms had cleared and the *Demodex* density was normalized on the preauricular zone ($0 + 0$ D/cm^2). We concluded that the IPL may have killed the mites on the treated zones with release of their antigens and flare-up of the inflammation, or that the mites may still have persisted more deeply in the skin, in sufficient number to induce the inflammation. The standardized skin surface biopsy (SSSB1 + SSSB2) values are indicated on the figure. Part **a**, so also the zoom **b**, was reprinted from "Gallo RL et al. Standard classification and pathophysiology of rosacea: The 2017 update by the National Rosacea Society Expert Committee. J Am Acad Dermatol. 2018;78(1):148–55", 2018, with permission from Elsevier. Parts **c** and **d** were reprinted from "Forton FMN, De Maertelaer V. Erythematotelangiectatic rosacea may be associated with a subclinical stage of demodicosis. A case control study. Br J Dermatol. 2019; 181: 818–25", 2019, with permission from John Wiley and Sons

detect it, may go unnoticed, so that this entity is often underdiagnosed [56, 98, 102, 104]. To detect the follicular scales, the dermatologist must examine the skin from a distance of maximum 30 cm, with good tangential lighting; sometimes cleaning the skin with ether may be necessary to reveal the scales [97, 99, 101, 105]. Subjective complaints (sensation of burning, pruritus, dry skin, hypersensitive skin, irregular or rough skin) may also be present [7, 56, 57, 99, 101–103]. Nevertheless, despite the discreet symptoms (likely because of the mites' control over host immunity), *Demodex* densities in the skin of these patients are usually high [101–103], with values ranging from 7 to 61 D/cm^2 depending on the sampling method used and the population studied [56, 57, 106, 107], and reaching as much as 285 ± 12 D/cm^2 when the densities of two consecutive standardized skin surface biopsies (SSSBs) are summed [108, 109].

Over time, a more inflammatory stage of the disease may occur. Despite the local immunotolerance likely induced by the mite, the host immune system mounts a chronic, exaggerated, and not very effective response, resulting in the development of the papules and pustules of demodicosis [57, 104, 110], clinically represented by "rosacea-like demodicosis" and other variants (*Demodex* folliculitis/abscesses, demodectic prurigo, isolated inflammatory papule, follicular eczematids, demodectic post inflammatory pigmentation, and ocular demodicosis) [7, 8, 10, 56, 57, 72, 101–103, 111–122]. The exact prevalence of demodicosis is unknown, but is at least 1.5 times more frequent than PPR in dermatological consultations [56].

DEMODEX PROLIFERATION IN ROSACEA: A CONTINUUM PROCESS?

Most authors still consider that proliferation of the *Demodex* mite in patients with rosacea is a secondary event, an epiphenomenon or an aggravating factor in which the initial inflammation promotes the proliferation of *Demodex*, which then exacerbates the disease [25, 123–127]. However, multiple observations suggest that the *Demodex* mite may itself contribute to the early inflammatory process. Indeed, in histological studies, *Demodex* mites are found in 63% of cases with ETR, 85% to almost 100% of cases of PPR, and in 100% of hypertrophic forms of rosacea [31, 94]; they have also been identified in intradermal granulomas in 3–66% of patients with granulomatous rosacea [25, 128–131]. The mean facial density of the mite in patients with ETR is between the low density found in subjects with healthy skin and the very high density in those with demodicosis and PPR [95, 97, 132–135]. In an observational study, we observed an abnormally high *Demodex* density in about half of our patients with ETR (10/23 patients), with high variability in values showing that different patients had different degrees of *Demodex* proliferation [97]. As we took particular care not to include patients with discreet pityriasis follicularum in our ETR group, we concluded that ETR may be associated with non-visible *Demodex* proliferation, possibly corresponding to a subclinical stage of demodicosis [97].

As PPR is more often observed after ETR than the inverse among patients with both PPR and ETR [136], and as the *Demodex* mite may be responsible for the papules and pustules of rosacea, this suggests that ETR is a condition that promotes mite proliferation [97], via a mechanism that is still unclear. One hypothesis is that vasodilation increases skin temperature [27, 31, 94], potentially promoting parasite reproduction, but the temperature, although it may be increased during flushes [137], does not appear to be higher in the skin of patients with ETR than in those without [138].

While it likely induces immunotolerance, *Demodex* is also able to stimulate the immune system's defense reaction [34, 43, 57, 58, 104, 110]. It stimulates Toll-like receptor 2 (TLR2) [58], resulting in an increased production of LL-37, with the subsequent angiogenesis and inflammation that are described in rosacea [15–18]. This implies the existence of a vicious circle including ETR, mite proliferation, and inflammation [10].

Demodex proliferation therefore seems to contribute to the continuum process in rosacea across all phenotypes.

DEMODEX AND IMMUNOTOLERANCE: A ROLE FOR VEGF AND THOMSEN-NOUVEAU ANTIGEN (TN AG)?

The apparent effect of ETR on *Demodex* proliferation [27, 31, 94, 97] may be explained by the immunosuppressive properties of VEGF, which were recently described in tumor pathology. VEGF inhibits maturation of dendritic cells, induces accumulation of immunosuppressive cells, such as regulatory T cells, and inhibits the migration of T lymphocytes to the tumor, thus favoring tumor cell escape from immune system surveillance [139, 140]. In rosacea, VEGF and its receptors, VEGF-R1 and VEGF-R2, are expressed not only by the epidermis, as in normal skin, but also by dermal infiltrating leukocytes (including lymphocytes, macrophages, and plasma cells) [141]. Moreover, accumulation of regulatory T cells has been observed [36, 38], as in demodicosis [142]. This suggests that, as in tumoral processes, VEGF may induce T cell exhaustion in rosacea and, through collaboration with tolerogenic dendritic cells, may favor the initial proliferation of the mite during the development of ETR (Fig. 2). The fact that *Demodex* densities can be normal in as many as half the patients with ETR [97] may be explained by a time lag between the immunomodulatory and pro-angiogenic effects of VEGF.

In addition to the direct action of VEGF on the maturation of dendritic cells [139], tolerogenic dendritic cells may be induced by several mechanisms. Initially, their production may be stimulated by high levels of thymic stromal lymphopoietin (TSLP) observed in sebaceous gland-rich zones of the healthy skin, where it induces an immunotolerant milieu for commensal microbes [36]: this cytokine exists in two forms, a long (inflammatory) and a short (tolerogenic) isoform [143] and is produced by keratinocytes in response to microbial products, physical injury, or inflammatory cytokines [144].

Tolerogenic dendritic cells may also be induced by vitamin D_3 and/or endogenous glucocorticoids. Indeed, in rosacea, TLR2 stimulates the enzyme responsible for the second hydroxylation of vitamin D_3 in keratinocytes, which initiates the inflammatory cascade [18] and promotes innate immunity [17]. But vitamin D_3 also inhibits adaptive immunity: exogenous treatment with vitamin D_3 promotes tolerogenic dendritic cells and increases expression of PD-L1 in dendritic cells, thus suppressing T cell proliferation [145]. The combination of dexamethasone and vitamin D_3 is an even more potent inducer of tolerogenic dendritic cells [145]. Furthermore, in ETR, abnormal glucocorticoid endogenous synthesis has been observed [146].

Tolerogenic dendritic cells may also be induced by the *Demodex* mite: because *Demodex* expresses the Tn Ag [147], it is possible that the mite could use this to induce immunotolerance for its own benefit (Fig. 3). Indeed, the Tn Ag is a precursor of the tumor Thomsen–Friedenreich (T) antigen [147]. These two antigens are tumor-associated glycan structures, and high expression levels are correlated with poor prognosis and an increased ability of the tumor to metastasize [147, 148]. Recently, it was shown that Tn Ag is recognized by the macrophage galactose-type lectin receptor (MGL) of the dendritic cell, which, on contact with it, becomes tolerogenic [148], inducing T cell exhaustion [149–152] (Fig. 3). After stimulation of its TLR2, producing a slight pro-inflammatory reaction, the dendritic cell usually also produces interleukin-10 (IL-10) as a natural feedback loop to prevent excessive inflammation. When its MGL receptor is also stimulated, this production of IL-10 is markedly increased, the two receptors working synergistically [151]. IL-10 is thought to play a pivotal role in blocking the metabolic switch to glycolysis (which is linked to immunogenic functions) and stimulating the expression of inhibitory receptors on dendritic cells (including PD-L1) and cytokines that induce transformation to tolerogenic dendritic cells [145] (Fig. 3). As Demodex mites express the Tn Ag [147], these immune reactions may also occur after contact of mite Tn Ag with dendritic cells.

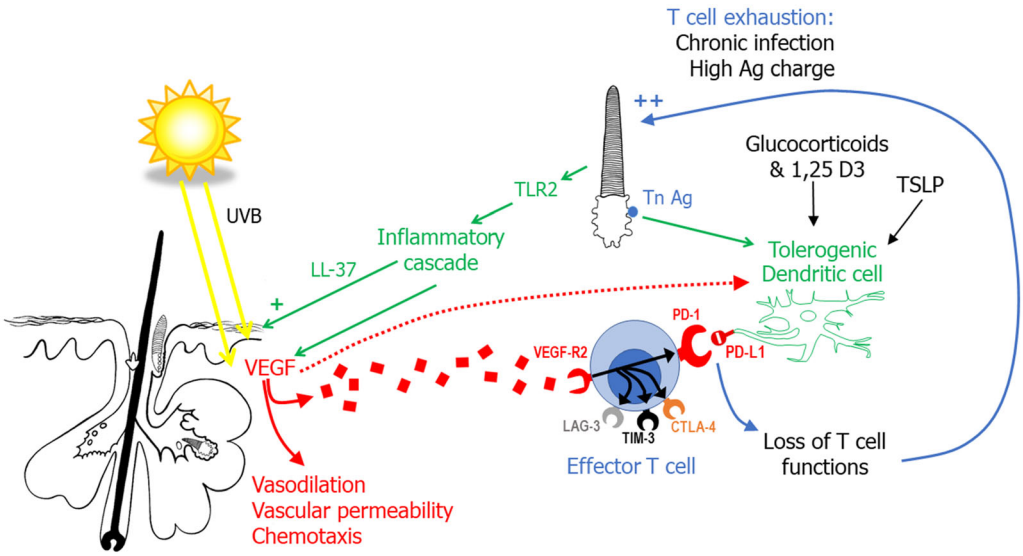

Fig. 2 Schematic of hypothesis that vascular endothelial growth factor (VEGF) may promote T cell exhaustion and therefore *Demodex* proliferation, by analogy with what happens with tumors. This figure assembles information from tumor pathology, from studies on *Demodex* and rosacea, and from the hypothesis formulated in Fig. 3. In tumor pathology, it is known that tumors secrete VEGF, which favors their development through its pro-angiogenic properties, but also by favoring T cell exhaustion: when VEGF is bound by the VEGF receptor (VEGF-R2) present on $CD8^+$ cytotoxic T lymphocytes, inhibitory receptors, such as programmed cell death 1 (PD-1), cytotoxic T lymphocyte antigen (CTLA-4), T cell immunoglobulin and mucin 3 domain (TIM-3), or lymphocyte activation gene 3 protein (LAG-3), are expressed on the lymphocyte surface [140]. When these receptors bind to their ligands, expressed by the tumors, this causes loss of lymphocyte function, with accumulation of regulatory T cells, reflecting T cell exhaustion [140]. In the skin, VEGF is produced by keratinocytes and fibroblasts under ultraviolet (UV) B induction [237, 238] and is increased in the dermis in rosacea, both in erythematotelangiectatic rosacea (ETR) and papulopustular rosacea (PPR) [26]. VEGF may play the same role in rosacea as in tumor pathology and collaborate with the tolerogenic dendritic cells to induce T cell exhaustion. The PD-1 receptor, induced on the effector T cell surface by its synapse with VEGF, binds to the programmed death ligand 1 (PD-L1), expressed on the surface of tolerogenic dendritic cells: this synapse then causes a loss of T cell function [145]. Tolerogenic dendritic cells may be induced by the mite (Fig. 3), thymic stromal lymphopoietin (TSLP) [36], vitamin D_3 (1,25 D3) and/or glucocorticoids [145, 149], and production is also favored by VEGF [139]. The *Demodex* mite activates a Toll-like receptor 2 (TLR2) pathway immune response [58], which induces increased production of the cathelicidin peptide, LL-37, and subsequent angiogenesis and inflammation [15, 18]. As LL-37 stimulates the activity of endothelial cells after UV exposure and may lead to increased sensitivity to UVB radiation [20, 21], theoretically, *Demodex* mites may also contribute to a higher sensitivity of the skin to UVB. This suggests a vicious circle that includes mite proliferation, TLR2, LL-37, sensitivity to UVB, and VEGF, providing a physiopathogenic link between ETR and PPR

ROSACEA: CHRONIC *DEMODEX* INFECTION WITH T CELL EXHAUSTION?

Demodex therefore likely induces tolerogenic dendritic cells via its Tn Ag, for its own survival (Fig. 3). It also induces a defensive, immunogenic immune reaction aimed at eliminating the mite, but probably succeeds in diverting this for its own benefit, by using VEGF and induced tolerogenic dendritic cells to cause T cell exhaustion (Fig. 2), as has been reported in dogs [153]. As some of the accumulated T cells will have lost their normal function [36, 38], the inflammatory reaction is likely to be insufficient to kill the mites, leading to a chronic infection

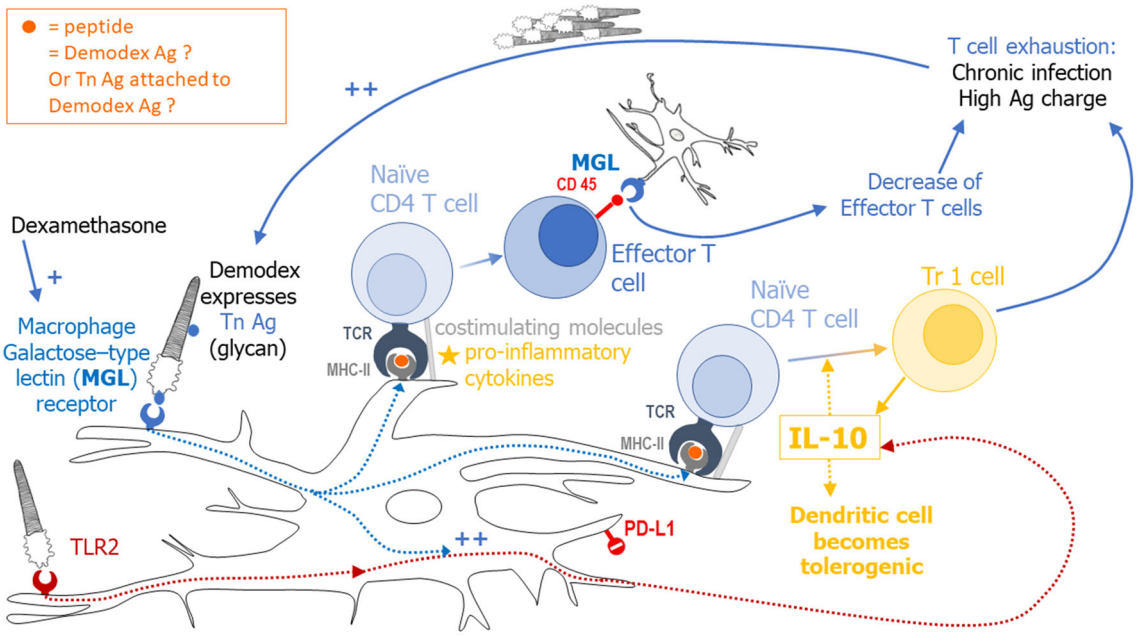

Fig. 3 How *Demodex* may manipulate the host immune system via its Tn Ag to induce dendritic cell immunotolerance. This schematic figure assembles information from immunological studies on dendritic cells and from immunohistological studies on *Demodex* and rosacea. Immunological studies have shown that when dendritic cells connect with the Thomsen-nouveau antigen (Tn Ag), through its macrophage galactose-type lectin receptor (MGL), the cells migrate towards the draining lymph node, where they initiate adaptive immunity [149]. The dendritic cells interact with naïve T cells to induce immunotolerance: a peptide Ag (small orange circle) with the major histocompatibility complex (MHC) type II is presented to the T cell receptor (TCR) of the naïve T cell, together with co-stimulation molecules (gray bar). If the dendritic cell also secretes pro-inflammatory cytokines (yellow star), the Ag presentation transforms the naïve T cell into an effector T cell expressing CD45. This interacts again with the MGL receptor of the dendritic cell [150], inducing loss of the functions of the effector T cell (decreasing proliferation, reducing production of inflammatory cytokines, and increasing apoptosis) [151]. If, instead of pro-inflammatory cytokines, there is interleukin (IL-10), contact with the naïve T cell results in its transformation into a Tr1 lymphocyte (with immunosuppressive functions), which in turn produces more IL-10 [148, 152]. The production of IL-10 by the dendritic cell after stimulation of its TLR2 is strongly increased when the MGL receptor is also stimulated [151]. IL-10 is thought to induce tolerogenic transformation of the dendritic cell and to stimulate the expression of inhibitory receptors (including programmed death-ligand 1 (PD-L1)) [145]. As *Demodex* mites express the Tn Ag [147], these immune reactions may also occur after contact of mite Tn Ag with dendritic cells. The peptide Ag (small orange circle) presented by the dendritic cells to the naïve T cell may be another *Demodex* Ag (exocuticle [239], proteases [167], its endosymbiont *Corynebacterium kroppenstedtii* [240], etc.) or the Tn Ag attached to a peptide *Demodex* Ag. The *Demodex* mite has also been shown to stimulate TLR2 [58], expression of which is increased in rosacea [18]. Dexamethasone treatment upregulated MGL expression on dendritic cells [149], and, in ETR, abnormal endogenous glucocorticoid synthesis has been observed [146]

with persistence of a high antigenic load. This hypothesis places the dendritic cell, together with the mite, at the heart of the pathophysiology of rosacea and, because of the existence of different types of dendritic cells (polymorphism of dendritic cell genes), may explain the differences in individual susceptibilities to *Demodex* antigens, and thus some of the genetic influence, as in inflammatory bowel disease [154].

DIAGNOSTIC CONFUSION

The potential role of the *Demodex* mite in the development of rosacea and the multiple similarities between demodicosis and rosacea lead to considerable diagnostic confusion. More work needs to be done to reach agreement on the diagnosis and relationship among demodicosis, ETR, and PPR, potentially leading to a consensus that they are all part of the same entity (Figs. 4 and 5) [8, 10, 155].

Pityriasis Folliculorum and ETR

When pityriasis folliculorum is associated with flushes, erythema, and/or telangiectasia, these obvious vascular symptoms may overshadow the discreet follicular scales of pityriasis folliculorum which are more difficult to identify (Figs. 1c, 5), thus potentially leading to a misdiagnosis of ETR based on the presence of persistent erythema. This diagnostic confusion may explain the unusually high *Demodex* densities observed in some studies of patients with ETR, similar to those observed in patients with PPR [29, 156]. Some patients with pityriasis folliculorum (with subclinical, subtle, or even obvious follicular scales) were probably misdiagnosed as ETR. Some authors may also have confused follicular scales with dry skin, with some even talking about two subtypes of ETR—scaly and not scaly ETR [157]—instead of the more likely diagnosis of pityriasis folliculorum. Others have suggested that this dry and rough aspect, with the possibility of fine follicular scales, is a characteristic of ETR [158]. This possible confusion highlights the importance of careful skin examination by a dermatologist experienced in the diagnosis of demodicosis.

The ROSCO consensus specifies that demodicosis must be excluded before diagnosing rosacea, but without specifying how this should be done [13]. Interestingly, on the photo selected by the NRS to illustrate rosacea with only persistent erythema [6], there is evidence of the presence of follicular scales, suggesting that, according to the NRS, patients with ETR may have follicular scales, and thus pityriasis folliculorum (Figs. 1a, b, Fig. 4, Table 1). Moreover, as a secondary ocular manifestation of rosacea, the NRS included collarette accumulation at the base of the lashes [6], likely corresponding to the cylindrical dandruff/follicular scales described in ocular demodicosis [118].

Rosacea-Like Demodicosis and PPR

If *Demodex* mites induce the immune response that leads to the papules and pustules of PPR [8, 10, 29, 31, 34, 35, 56–58, 99, 108, 110, 111, 128, 132–134, 159–170], then rosacea-like demodicosis and PPR are probably two phenotypes of the same disease [10]. Indeed, the descriptions of demodicosis and rosacea seem to indicate two different approaches to the same condition (Table 1): their definitions cannot be compared because they are based on different criteria (etiological for demodicosis [101, 102] and clinical for rosacea [5, 6]); their symptoms are similar [10, 31] with no single criterion being specific for either and most patients presenting a mixture of characteristics that can be attributed to both of the entities [10]; they may occur successively in the same patient [10]; their histological characteristics are similar [31]; their *Demodex* densities are similar [10]; and both respond very well to acaricidal treatment, in terms of reduced *Demodex* densities and improved clinical symptoms [159, 160, 171, 172]. It is therefore increasingly difficult to defend the view that, in these two similar diseases, the exaggerated proliferation of parasites has a different role, causal in one and epiphenomenal in the other.

The *Demodex* mite is beginning to be accepted as one of the triggers that stimulates TLR2 at the start of the inflammatory cascade in rosacea [5, 14, 93, 123, 173], and as a marker of rosacea [174]: a clinical diagnosis of PPR may be confirmed by a diagnostic test based on the high *Demodex* density present in these patients, using two SSSBs taken from the same site. A superficial [SSSB1] *Demodex* density greater than 5 D/cm^2 OR a deep [SSSB2] *Demodex* density greater than 10 D/cm^2 enabled confirmation of a diagnosis of PPR (or demodicosis) with a sensitivity of 98.7% and a specificity of 95.5% [108].


a
Pityriasis folliculorum (PF) type DEMODICOSES:
Follicular scales with or without persistent erythema
ETR according to NRS:
Persistent erythema
with or without follicular scales
Follicular scales without persistent erythema
Follicular scales with persistent erythema
=
Persistent erythema with follicular scales
Persistent erythema without follicular scales
b
Pityriasis folliculorum (PF) type DEMODICOSES
= clinical PF (follicular scales)
+ subclinical PF
(persistent erythema with high Demodex density without follicular scales)
ETR according to NRS:
Persistent erythema
with or without follicular scales
Subclinical PF with erythema
Clinical PF without erythema
Clinical PF with erythema
=
ETR
with follicular scales
ETR without follicular scales
c
Pityriasis folliculorum (PF) type DEMODICOSES
n= 455
= 445 clinical PF
+ 10 subclinical PF with erythema
According to NRS, ETR: n= 332
= 23 clinical ETR
without follicular scales (10+13)
+ 309 ETR with follicular scales
Subclinical PF with erythema
n=10
+++ +++
Clinical PF without erythema
n= 136
Clinical PF with erythema
=
ETR
with follicular scales
n= 309
« Pure »
ETR
n=13

◀ **Fig. 4** Overlap of the clinical entities pityriasis folliculorum (PF) and erythematotelangiectatic rosacea (ETR). **a** Schematic conceptualization of the overlap between PF and ETR. Most cases of "ETR according to the National Rosacea Society (NRS) definition" have follicular scales. Patients with the pure vascular form of rosacea (without increased *Demodex* density/follicular scales) or pure demodicosis (without persistent centrofacial erythema) are encountered less frequently. "PF with persistent erythema" and "ETR with follicular scales" are likely the same entity, with two possible modes of entry: (1) ETR, likely through VEGF, favors mite overproliferation [8, 10, 25, 27, 31, 57, 93–97], which in turn results in increased erythema as a result of inflammation; and (2) other factors favoring mite proliferation give rise initially to PF without erythema, but persistent erythema then develops as a result of the inflammation caused by the mites. **b** Some patients have persistent erythema without visible follicular scales, but nevertheless have high *Demodex* densities (Dd): they are considered to have subclinical demodicoses/subclinical pityriasis folliculorum, and so are included into the demodicosis group. **c** As a practical example of the schema in **b**, we provide the data from the 445 patients clinically diagnosed with pityriasis folliculorum and 23 patients clinically diagnosed with ETR from our recent studies [97, 99, 108]. According to the NRS definition, 332 of these 468 patients would have been diagnosed as ETR, of whom 309 (93%) had follicular scales. Clinically, among these 332 patients, we diagnosed only 23 patients as having ETR (without follicular scales) and 309 as having pityriasis folliculorum; only 13 had "pure" ETR, without *Demodex* proliferation detected. Patients with persistent erythema and high Demodex density (i.e., subclinical PF with erythema or clinical PF with erythema/ETR with follicular scales) were the most numerous (319/468 = 68%). In real life, the proportion of patients with ETR without follicular scales is nevertheless certainly higher ("+++") because in our studies these patients were only included when we had time to perform the SSSB, whereas all patients with follicular scales were included [97, 99, 108]

CURRENT TREATMENTS FOR VASCULAR SYMPTOMS OF ROSACEA: ACARICIDAL EFFECTS?

Light Therapies

Light-based treatments with (long) pulsed dye laser (PDL), neodymium-doped yttrium aluminum garnet (Nd:YAG) laser, and intense pulsed light (IPL) target oxyhemoglobin in the vascular system [155], but may also have acaricidal actions in rosacea. PDL, which had earlier been reported to have limited value for treatment of papulopustules [175], was recently shown to decrease their number [176]; this action, as well as its effect on erythema, tended to be more marked when ivermectin was given topically at the same time [176]. Nd:YAG laser also acts on papulopustules but is more effective in ETR than in PPR [177]. These actions on the papulopustules may result from a potential acaricidal effect as a result of increasing skin temperature [178] and, for the Nd:YAG laser, by destruction of the follicular unit [177]. One study suggested that these therapies may therefore be used not only in ETR but also in demodicosis [178], although these findings need to be confirmed. Coagulation necrosis of the *Demodex* mites has been observed after IPL treatment; nevertheless, the mites recolonized the skin 1 month after the second treatment [179]. IPL reduced the risk of recurrence of PPR after oral acaricidal treatment [180, 181], which also supports a facilitating role of the vascular background on the proliferation of mites.

Paradoxically, if these light-based therapies are applied from the outset of treatment, they may cause an exacerbation of rosacea, probably related to the mass death of many *Demodex* mites, although not all, because some are still observed after treatment [182] (Fig. 1e, f). Patients should therefore be treated first with an acaricidal treatment, and then possibly by laser or IPL treatment [182].

Vasoconstrictors

Topical vasoconstrictors, such as α-blockers (brimonidine, oxymetazoline) [183, 184], are used to decrease vasodilation of the dermal capillaries for a limited time, and act mainly on erythema but not on telangiectasia [2]. They are mainly recommended after light therapy, when this is not completely successful [185]. To our knowledge, these treatments have no activity against the *Demodex* mite.

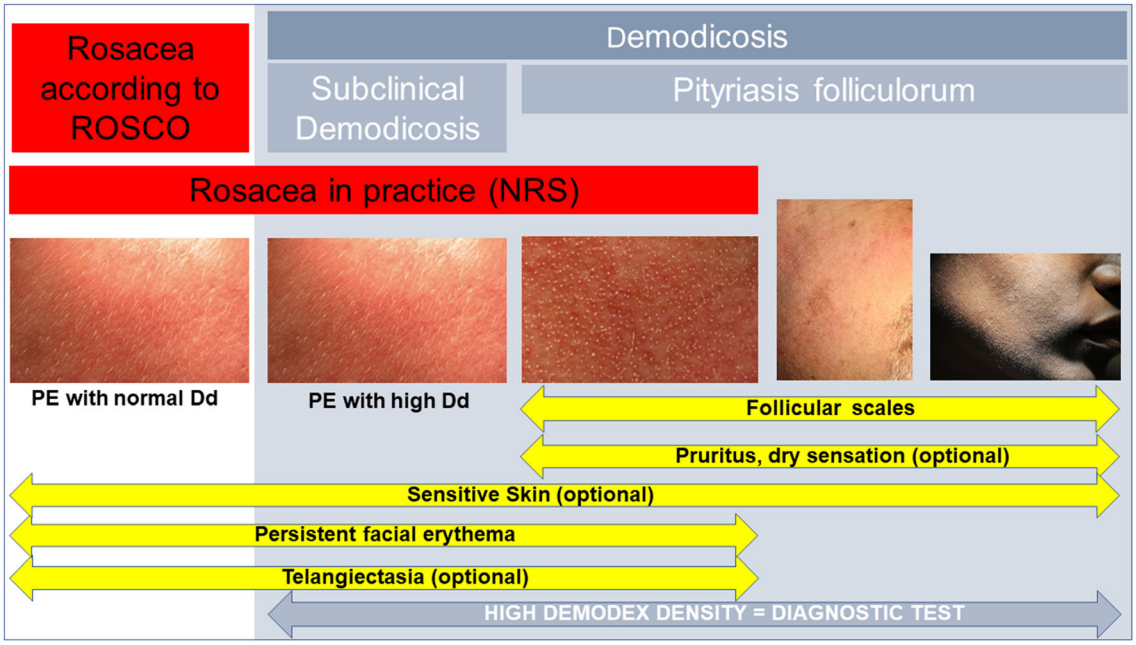

Fig. 5 Rosacea with only vascular symptoms and pityriasis folliculorum: overlaps and ambiguities in diagnostic criteria. According to the ROSacea COnsensus (ROSCO) panel, demodicosis must be excluded before making a diagnosis of rosacea [13], but it was not specified how this should be done; however, it can be assumed that the *Demodex* density must be normal. The latest National Rosacea Society (NRS) consensus seems not to take follicular scales into consideration to include or exclude a diagnosis of rosacea: the consequence is that patients with pityriasis folliculorum (and subclinical demodicosis) with vascular symptoms may therefore be (mis)diagnosed as having rosacea with only vascular symptoms (ETR). However, pityriasis folliculorum is not always associated with ETR (the two photos on the right, on white skin and black skin). Of note, the photograph illustrating "Persistent erythema (PE) with normal *Demodex* density (Dd)" is the same as that of subclinical demodicosis "PE with high Dd" to stress that the clinical appearance/phenotypes of these conditions are identical. The patient with pityriasis folliculorum on black skin has provided written consent for publication

ACARICIDAL TREATMENTS

Case Reports

Multiple molecules have been reported to reduce the number or density of *Demodex* mites and improve or cure symptoms of demodicosis and rosacea in case studies.

Topical treatments have included sulfur or selenium (di)sulfide [75, 81, 101, 102, 105, 114, 115, 186–188], lindane (currently prohibited) [73, 74], yellow mercury oxide [189], malathion [72], metronidazole [190, 191], permethrin [71, 73, 192], pilocarpine [193, 194], benzyl benzoate [195], and combinations of some of these treatments (also with crotamiton) [103, 196–199]. Oral treatments have included ivermectin (alone [200] or associated with topically administered crotamiton [201] or permethrin [202]) and metronidazole (alone [121, 203] or associated with topically administered metronidazole [197] or crotamiton [204]).

Some case reports in which these treatments were used also observed a clinical effect on lesions rich in *Demodex* but did not check that *Demodex* levels decreased after treatment [66–68, 70, 77, 120, 205–207].

Other case studies have reported no clinical effects of some of these treatments in demodicosis, e.g., permethrin [68, 203], crotamiton [70], topically administered [68, 70, 203] and

Table 1 Comparison between pityriasis folliculorum and erythematotelangiectatic rosacea (ETR), and between rosacea-like demodicosis and typical papulopustular rosacea (PPR)

	Pityriasis folliculorum	**ETR according to NRS definition**	**References**
Definition	Etiological	Clinical	[6, 101, 102, 105]
Cause	*Demodex* proliferation	Inflammatory reaction?	[6, 101, 102, 105]
Demodex density	Similar		[29, 109, 156]
Histology	Similar		[31]
Signs and symptoms	Similar, mixed forms are the most frequent		[6, 97], Fig. 4c
Main characteristic	Follicular scales	Persistent centrofacial erythema associated with periodic intensification by potential trigger factors	[6, 101, 102, 105]
Persistent erythema	Frequent	Always	[6], Fig. 4c
Follicular scales	Nearly always	Frequent	[99], Fig. 4c
Facial location	All	Central part	[6, 99, 101, 102, 105, 114]
Treatment	Acaricide	Light treatment, vasoconstrictors	

	Rosacea-like demodicosis	**PPR according to NRS definition**	**References**
Definition	Etiological	Clinical	[6, 102, 105]
Cause	*Demodex* proliferation	Inflammatory reaction? *Demodex* proliferation?	[6, 102, 105]
Demodex density	Similar, very high		[10]
Histology	Similar		[31]
Signs and symptoms	Similar, mixed forms are the most frequent		[10, 113]
Follicular scales	Similar		[99]
Skin	More dry	More greasy	[102, 105]
Facial location	Central and lateral parts	Central part	[6, 102, 105]

Table 1 continued

	Rosacea-like demodicosis	PPR according to NRS definition	References
	More unilateral	More bilateral	[71, 120, 201, 204, 207, 208]
Persistent erythema	Frequent	Always	[6, 102, 105]
Flushes/erythema	Less intense	More intense	[6, 102, 105]
Papules and pustules	More superficial and smaller	Deeper and larger	[102, 105, 113]
Pruritus	Usually considered more frequent in demodicosis		[70, 72, 74, 79, 241, 242, 99]
Evolution over time	The 2 forms may occur successively in the same patient		[10]
	Often after pityriasis folliculorum	More often after ETR than the reverse	[8, 57, 136]
Treatment	Acaricide	Anti-inflammatory, acaricide	

orally administered [204, 208] metronidazole, and orally administered ivermectin [121].

Experimental and Clinical Studies

Some authors have studied the survival time of the *Demodex* mite in vitro [209–213]. Tea tree oil and its isolated active component were shown to have considerable acaricidal activity [117, 210–212]; although they have mainly been used in *Demodex* blepharitis [117, 214, 215], they therefore also seem to be a promising treatment to kill the mites in the facial skin [216]. A relatively crude in vitro experiment using different concentrations of metronidazole showed that *Demodex* mites survived at a concentration of 1 mg/ml, a level that cannot be obtained in vivo [213]: the authors therefore suggested that metronidazole may act not directly in vivo but via one of its metabolites [217]. In a randomized clinical study comparing six topical treatments, we found that metronidazole had no acaricidal activity as measured using *Demodex* densities [218]. In a single-blind randomized study comparing an oral metronidazole-based treatment with a treatment based on oral ornidazole administration (a metronidazole analogue with a longer half-life), ornidazole was more effective than metronidazole (in terms of Demodex counts and clinical symptoms) and associated with fewer relapses [180].

In a randomized study, oral metronidazole treatment increased the acaricidal action seen with orally administered ivermectin [219], although the acaricidal effects of orally administered ivermectin have never been confirmed, especially over the long term, likely because the treatment itself is very short (2 weeks).

An acaricidal effect of permethrin cream applied twice daily was reported in two controlled studies [220, 221], and this treatment was proposed as a valuable option in a recent review [222].

Our randomized clinical study comparing six topical treatments reported that benzyl benzoate had marked acaricidal action and crotamiton moderate action [218]. We recently demonstrated the short- and long-term actions

of benzyl benzoate (with crotamiton) on *Demodex* density and on clinical symptoms in a real-life study [159, 160]. In our practice, we successfully used benzyl benzoate (with crotamiton) cream for more than 20 years; however, the development of ivermectin has provided an effective alternative [172, 223, 224] with better tolerance and this is now our treatment of choice. Indeed, ivermectin was approved by the US Food and Drug Administration and the European Medicines Agency in 2014–2015 as a topical anti-inflammatory treatment for PPR as a 1% once daily application [225]. Ivermectin quickly became established, together with azelaic acid, as a first-line treatment for PPR in mild rosacea [2, 183, 184, 223, 226–229], and combined with orally administered doxycycline in moderate to severe rosacea [173, 230, 231]. However, its efficacy may, at least in part, be explained by its other important property: acaricidal effects against the *Demodex* mite [172, 173, 225, 228, 230, 232–236]. Indeed, two other acaricidal treatments which have no known anti-inflammatory properties also improve clinical symptoms of rosacea: permethrin (5% applied twice daily) improved the vascular component of rosacea (erythema [221], telangiectasia [220]), and benzyl benzoate was shown to be an effective treatment for PPR [159, 160]. These observations provide indirect support for the role of the mite in PPR, as already suggested by numerous other studies [8, 10, 29, 31, 34, 35, 56–58, 99, 108, 110, 111, 128, 132–134, 161–170].

Acaricidal Treatment for ETR?

Topical acaricidal treatment is certainly the most appropriate treatment for patients with pityriasis folliculorum with vascular symptoms/ETR with follicular scales: this therapy kills the mites and decreases subsequent inflammation and associated persistent erythema. In our experience, topical acaricidal treatment leads to disappearance of follicular scales [159, 160] as well as subjective complaints, such as burning sensation and hypersensitive skin. If only mild erythema is present, it may completely, or almost completely, resolve after eradication of the mites, so that supplementary treatment (e.g., light therapy) may not be needed. More severe erythema generally just decreases a little in intensity, but the acaricidal treatment is nevertheless useful because subsequent light treatment may be better tolerated, without the potential for flare-ups [182].

Acaricidal treatment may also prevent the immunotolerance induced by the mite, and its subsequent overproliferation and ultimately the development of the papulopustules of rosacea, although further study is needed to confirm this hypothesis.

Because the distinction between ETR and pityriasis folliculorum is often not made, and even experienced dermatologists may miss subclinical demodicosis in about 40–50% of cases, a pragmatic approach may be to start treatment of all patients diagnosed with "ETR" with an acaricide (because many of them will have undiagnosed pityriasis folliculorum) for 2–4 months. A more scientific approach would be to measure the *Demodex* density in all patients with suspected ETR and start treatment with an acaricide only when the *Demodex* density is high and until the *Demodex* density normalizes (followed by a maintenance therapy). This can be managed easily in the clinic using two consecutive SSSBs as discussed earlier [108].

If future experimental studies confirm that VEGF collaborates with the mite to induce immunosuppression in rosacea, thus favoring *Demodex* proliferation, acaricidal treatment would then be clearly indicated in any patient with vascular symptoms of rosacea, and may contribute to prevent further evolution of ETR.

CONCLUSION

The interactions among VEGF, *Demodex*, and the immune system need further exploration and the nosology of rosacea would then need to be adapted accordingly. The effectiveness of treating any patient with ETR first with an acaricidal cream needs to be assessed in prospective controlled clinical trials with long-term follow-up. Currently, learning to distinguish patients with pityriasis folliculorum from those with

isolated ETR is crucial so that they can be managed appropriately with an acaricidal cream.

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