INTRODUCTION

Telogen effluvium (TE) is a type of acquired, diffuse alopecia that occurs due to an abnormal shift of scalp hair follicles from anagen to telogen, leading to premature shedding of hair. While data on the epidemiology of TE is limited, it remains one of the most common forms of nonscarring hair loss for which patients present for clinical evaluation. TE can occur at any age, and for reasons that are unclear women are more likely than men to present for the evaluation of acute TE. The major clinical finding in TE is an acute or chronic decrease in scalp hair density.

Acute TE involves hair loss 2–4 months following a stressful inciting event. A wide variety of inciting factors have been associated with the induction of TE based on clinical observations. These factors include but are not limited to thyroid dysfunction, nutritional deficiencies, rapid weight loss, significant blood loss, childbirth, a myriad of prescription medications, and any physiologic or psychological stressor. However, the specific etiology of hair loss in TE still remains unclear. Several studies using mouse models have implicated a neuroimmunologic mechanism to account for hair loss in TE. Implicated specifically at the distal end of the “brain-hair follicle” axis.

ABSTRACT

Background: Telogen effluvium (TE) is a type of acquired, diffuse alopecia that occurs due to an abnormal shift of scalp hair follicles from anagen to telogen, leading to premature shedding of hair. Previous studies have suggested the existence of a neuroimmunologic “brain-hair follicle” axis, in which mast cells have been implicated as an important link between the nervous system and immunologic system. Objective: The current study sought to investigate the role of mast cell presence and mast cell degranulation in the pathogenesis of TE. Materials and Methods: Mast cells were counted using Giemsa and tryptase immunohistochemical stains in scalp biopsy specimens with the pathologic diagnosis of TE (TE, n = 10), alopecia areata (AA, n = 7), and androgenic alopecia (ANDRO, n = 9). Results: We found significant (P < 0.001) group-level differences between the mean mast cell counts per high-power fields for each type of alopecia studied. Tukey post hoc analysis showed the mean mast cell count for TE to be significantly larger than AA for both Giemsa (P = 0.002) and tryptase (P = 0.006); significantly larger than ANDRO for both Giemsa (P < 0.001) and tryptase (P < 0.001); and significantly larger when compared to normal scalp skin for both Giemsa (P < 0.001) and tryptase (P < 0.001). No significant difference of mean mast cell counts was observed for AA compared to ANDRO for Giemsa (P = 0.373) or tryptase (P = 0.598) stains. Conclusion: Our findings suggest that mast cells could play a role in mediating stress-induced hair loss seen in TE.

Key words: Alopecia areata, mast cells, telogen effluvium

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are mast cells which have been shown to modulate follicular cycling during times of stress.\textsuperscript{[12,13]}

The current study sought to investigate the role of mast cell presence and mast cell degranulation in the pathogenesis of TE compared to two other nonscarring alopecias: Alopecia areata (AA) and androgenic alopecia (ANDRO). Identifying differences in mast cell presence and activity between these three alopecias may further elucidate their pathogeneses and provide insight into possible therapeutic interventions for these conditions.

**MATERIALS AND METHODS**

**Case selection**

The approval for this study was obtained from the Saint Louis University Office of Research Administration. Our dermatopathology laboratory information system was searched for scalp biopsy specimens with a final pathologic diagnosis of TE, AA, or ANDRO from 2006 to 2011. Formalin-fixed and paraffin-embedded tissue was obtained from the dermatopathology laboratory at Saint Louis University in St. Louis, MO. Ten cases of TE, seven cases of AA, and nine cases of ANDRO were obtained and reviewed by a board-certified dermatopathologist (CIV) for diagnostic accuracy and inclusion in the study. Eight elliptical scalp excision specimens for benign nevi were selected, and the tips of these excision specimens were used as normal skin controls.

**Calculating the number of mast cells in each type of alopecia**

Mast cells were counted using 4-µm transverse sections with specific attention made to highlight the areas of greatest inflammation. Follicular and nonfollicular mast cells were included in the study. All specimens were stained with the histochemical marker Giemsa (Artisan Giemsa Stain Kit; Dako Denmark A/S, Denmark), which provides metachromatic staining of mast cell granules, and the immunohistochemical marker tryptase (Monoclonal mouse antihuman mast cell tryptase; Dako, Denmark A/S, Denmark), a main mast cell constituent, for identification of mast cells within each specimen.\textsuperscript{[14]} Alopecia areata cases were reviewed by the dermatopathology fellow (NA) and a board-certified dermatopathologist (CIV) who used three high-power fields (HPF) (×40) to count the number of mast cells within each specimen.\textsuperscript{[14]} Alopecia areata cases were counted using 4-µm transverse sections with specific attention made to highlight the areas of greatest inflammation. Follicular and nonfollicular mast cells were included in the study. All specimens were stained with the histochemical marker Giemsa (Artisan Giemsa Stain Kit; Dako Denmark A/S, Denmark), which provides metachromatic staining of mast cell granules, and the immunohistochemical marker tryptase (Monoclonal mouse antihuman mast cell tryptase; Dako, Denmark A/S, Denmark), a main mast cell constituent, for identification of mast cells within each specimen.\textsuperscript{[14]} Alopecia areata cases were reviewed by the dermatopathology fellow (NA) and a board-certified dermatopathologist (CIV) who used three high-power fields (HPF) (×40) to count the number of mast cells within each specimen. The three counts were averaged, and the mean number of mast cells per HPF for each specimen was used to calculate an overall total mean/HPF for TE, AA, and ANDRO. The same protocol was used for controls of normal scalp skin.

**Statistical analysis**

One-way analysis of variance (ANOVA) was used to determine statistically significant difference between the three group mean mast cell counts. Tukey post hoc analysis was then employed to calculate the statistical significance of mean mast cell counts between each group and each stain. Student’s t-test was used to compare the mean mast cell counts using Giemsa versus tryptase for each type of alopecia studied.

**RESULTS**

**Telogen effluvium mast cell counts**

A total of ten specimens of previously diagnosed TE [Figure 1] were examined. When using Giemsa for the identification of mast cells, the calculated mean number of mast cells per HPF for individual TE specimens ranged from 18 to 53 with a total mean for all specimens of 34.70 and a standard deviation (SD) of 12.82. When using tryptase for the identification of mast cells, the calculated mean number of mast cells per HPF for individual TE specimens ranged from 21 to 74 with a total mean for all specimens of 52.90 and a SD of 16.80 [Table 1].

**Alopecia areata mast cell counts**

A total of seven specimens of previously diagnosed AA [Figure 2] were examined. When using Giemsa for the identification of mast cells, the calculated mean number of mast cells per HPF for individual AA specimens ranged from 2 to 31 with a total mean for all specimens of 15.14 and a SD of 10.62. When using tryptase for the identification of mast cells, the calculated mean number of mast cells per HPF for individual AA specimens ranged from 8 to 31 with a total mean for all specimens of 24.33 and a SD of 8.83.
from 11 to 40 with a total mean for all specimens of 30.71 and a SD of 10.75 [Table 1].

**Androgenic alopecia mast cell counts**

A total of nine specimens of previously diagnosed ANDRO were examined. When using Giemsa for identification of mast cells, the calculated mean number of mast cells per HPF for individual ANDRO specimens ranged from 1 to 18 with a total mean for all specimens of 8.11 and a SD of 5.35. When using tryptase for the identification of mast cells, the calculated mean number of mast cells per HPF for individual ANDRO specimens ranged from 7 to 34 with a total mean for all specimens of 24.33 and a SD of 8.83 [Table 1].

**Normal scalp skin controls (CONTROL) mast cell counts**

A total of eight normal scalp skin specimens were examined. When using Giemsa for the identification of mast cells, the calculated total mean number of mast cells per HPF for all specimens \( (n = 8) \) was 11.63 with a SD of 2.97. When using tryptase for the identification of mast cells, the calculated total mean number of mast cells per HPF for all specimens \( (n = 8) \) was 20.50 with a SD of 6.97 [Table 1].

**Statistical analysis**

One-way ANOVA demonstrated statistically significant group-level differences [Figure 3] in the mean mast cells counts across the three types of alopecia in both Giemsa and tryptase as well as for TE compared to normal scalp skin controls [Table 1]. In addition, Student’s t-test analysis showed that within each type of alopecia, the tryptase staining mast cell mean was higher than the Giemsa staining mast cell mean.

**DISCUSSION**

TE is a type of acquired, diffuse, nonscarring alopecia which involves hair loss 2–4 months following a stressful inciting event. The relationship between psychoemotional stress and hair loss was first elucidated by Selye in 1950. Selye’s observation led to multiple investigations of the link between stress and sudden hair loss, many of which date back to the 1950s. Although a medically benign condition, alopecia can have a significant negative impact on one’s self-image, leading to a decreased quality of life. Despite the fact that an association between hair loss and stress is readily acknowledged by patients and physicians, a clear-cut pathophysiologival mechanism explaining the connection between psychoemotional stress and hair loss in humans remains to be demonstrated. Our analysis of scalp biopsy specimens demonstrated that TE specimens have a significantly higher number of mast cell counts per HPF compared to normal control skin, AA, and ANDRO specimens, indicating that mast cells could play a role in the pathophysiology of hair loss in TE.

In an attempt to further elucidate the pathophysiology of stress-induced hair loss, Arck et al., proposed a substance P(SP)-dependent neuroimmunological “brain-hair follicle” axis. Following the induction of a stressful event, the hypothalamic-pituitary-adrenal (HPA) stress response axis
Grace, et al.: Mast cells in telogen effluvium

**Figure 3:** Mean number of mast cells (+standard error mean) per high-power field in telogen effluvium, alopecia areata, androgenic alopecia and normal (CONTROL) scalp biopsies. Tukey post hoc analysis showed the mean mast cell count for the telogen effluvium group to be significantly larger than androgenic alopecia (Giemsa, *P = 0.002; Tryptase, *P = 0.006) and CONTROL (Giemsa and Tryptase, **P < 0.001) and also showed the mean mast cell count for telogen effluvium to be significantly larger than alopecia areata (Giemsa and Tryptase, **P < 0.001)

is activated. Many well-studied hormones that are released along the HPA axis, such as corticotropin-releasing hormone, adrenocorticotropic hormone, and glucocorticoids, have been associated with hair loss in mice. In addition, increased HPA axis tone is thought to stimulate SP release from sensory nerve fibers in the skin, which densely innervate scalp hair follicles. Locally released SP has been shown to induce hair follicle keratinocyte apoptosis by activating macrophages and mast cells to release several hair growth inhibiting bioactive molecules such as tumor necrosis factor-α, interleukin-1, and proteases. Furthermore, neurokinin-1 receptor (NK-1R) and SP knockout mice have been shown to be resistant to stress-triggered premature induction of catagen and hair follicle apoptosis, which suggests that mast cells express NK-1R and that the cross-talk between SP and NK-1R on mast cells is essential in mediating stress-related hair loss. Nerve growth factor (NGF) has also been shown to promote outgrowth of SP+ nerve fibers as well as degranulate mast cells downstream of SP. The administration of NGF-neutralizing antibodies following a stressful stimulus has been shown to abrogate premature onset of catagen, the number of perifollicular mast cells, and hair-follicle keratinocyte apoptosis in murine skin.

Several murine studies indicate that mast cells play a distal, intermediary role in stress-induced hair loss, serving as a link between the nervous system and immunologic system, as evidenced by the proximity of mast cells and sensory nerve endings in both normal and inflamed skin. The relationship between dermal SP+ sensory nerve endings and mast cells may be bidirectional. SP degranulates mast cells inducing cytokine expression and NGF expression and selective release of proteases. The combination of proteases and NGF subsequently promotes survival and outgrowth of sensory nerve fibers through protease-activating receptors on the surface of sensory nerve endings. Thus, mast cells act as modulators of hair follicle growth and regression by means of SP and NGF. Mast cells have also been reported to play a major role in AA. Bertolini et al. found the number, degranulation, and proliferation of perifollicular mast cells as well as mast cell-CD8+ T-cell physical contacts to be significantly increased in human AA lesions compared to healthy control skin. They proposed the interaction of mast cells with CD8+ T-cells causes the mast cells to switch from an immune-inhibitory to a pro-inflammatory phenotype that causes the anagen hair follicle to lose its immune privilege by upregulating major histocompatibility complex (MHC) class I molecules and downregulating the expression of immune privilege guardian molecules such as transforming growth factor beta-1. In addition, Peters et al. showed that cultured human anagen hair follicles exposed to SP promoted catagen transformation and increased MHC class I expression, suggesting that the loss of immune privilege also plays a role in stress-induced hair loss.

By analyzing human scalp biopsy specimens, the results of the current study corroborate the putative “brain-hair follicle” axis and contribute to the growing body of murine model, in vivo evidence that stress-induced hair loss is a neuroimmunologic phenomenon with mast cells playing an essential role at the distal end of the cascade. Currently, systemic treatment with NK-1R antagonists anti-NGF neutralizing antibodies and topical minoxidil has been shown to reduce stress-induced hair loss in murine models. Physicians should not underestimate the emotional impact of hair loss as it can often provoke distress that is out of proportion to its objective severity. As the investigation into the “brain-skin connection” continues, it is essential for researchers and clinicians to understand the hair follicle as a peripheral target of a myriad of bioactive molecules involved in the stress cascade. Theoretically, any step or molecule implicated in this cascade could serve as a therapeutic target, and clinical trials are needed. Our results suggest that mast cells could be implicated in the pathogenesis of hair loss in patients diagnosed with TE and that modulation of either systemic or perifollicular mast cell activity could be a promising therapeutic approach for these patients.
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conflicts of interest

there are no conflicts of interest.

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