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Treatment of alopecia areata with squaric acid dibutylester

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Abstract Squaric acid dibutylester is a topical sensitizing agent utilized for the treatment of alopecia areata. The mechanism of action is not fully understood, but is believed to redirect the inflammatory response by invoking allergic contact dermatitis. Several studies have compared the efficacy of squaric acid dibutylester to other treatments vs placebo with favorable results. This contribution reviews the history of the topical sensitizing agent squaric acid dibutylester and discusses the mechanism of action, the use in alopecia areata, and the efficacy and safety of this therapeutic agent.

Introduction

The treatments for alopecia areata (AA) are numerous, and many of them are quite ineffective.[1] The lack of randomized double-blind controlled trials and the incidence of spontaneous hair regrowth render validating a therapy difficult. One mode of treatment involving the induction of an allergic contact dermatitis has been shown to elicit hair regrowth in AA.[2] Chemicals or substances that elicit an allergic contact dermatitis and immunity upon exposure are called topical sensitizers. With subsequent exposures the sensitizer should produce a T-cell memory inflammatory response. Squaric acid dibutylester (SADBE) is a sensitizer found to induce hair growth in patients with AA in 1980.[3] SADBE is believed to elicit an inflammatory reaction to disrupt the autoimmune pathogenesis of the disease. Many studies have shown SADBE to be superior to placebo and other treatment options. As no needles or daily oral medication is employed with this regimen, SADBE can be used as a primary treatment for severe refractory AA in adults and children. This contribution reviews the use of SADBE, a topical sensitizing agent, in the treatment of AA. The mechanism of action, its use in the treatment of AA, and the efficacy and safety of SADBE are summarized.

Alopecia areata

AA is an autoimmune condition characterized by nonscarring hair loss involving the scalp, body, and nails. The prevalence of AA in the United States is 0.1% to 0.2% with a lifetime risk of 1.7%.[4,5] AA classically presents as one or more smooth, hairless patches on the scalp with preserved follicular ostia and no erythema. This localized presentation may progress to involve the entire scalp (alopecia totalis) and body and facial hair (alopecia universalis).

The exact cause of AA is unknown, but investigators have implicated a T-cell lymphocytes interaction with auto-antigens located in the hair follicles.[6] Various studies have demonstrated the immunologic influence on this condition. Skin transferred from the scalp of AA patients can regrow

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hair when grafted onto athymic nude mice. Severe combined immunodeficiency (SCID) mice can be induced to develop alopecic patches when injected with scalp-infiltrating CD4+/CD8+ T cells from a patient with AA (cite/Bolognia).

The hair follicle is an immune-privileged site with a low level of major histocompatibility complex (MHC) expression.[7] AA requires an interruption in this immune privilege, allowing infiltration of T cells into the hair follicle.[8] Triggering factors cause a predominantly CD8+ driven, Th1 T-cell response against hair follicles, resulting in acute hair loss. Certain proinflammatory molecules, including substance P and interferon γ (IFN-γ), have been shown to upregulate MHC class Ia expression in hair follicles. IFN-γ is the most potent inducer of MHC I expression in murine anagen hair bulbs *in vivo*, and similarly, IFN-γ was shown to accelerate the development of AA in genetically susceptible mice.[9,10] As IFN-γ is mainly produced by activated T lymphocytes, an activation of the immune system may precede the loss of immune privilege.[7]

Another theory is that AA is an autoimmune response against melanogenesis-related proteins. According to this theory, microtrauma, neurogenic inflammation, or microbial antigens leads to the production of proinflammatory cytokines, which are responsible for the breakdown of MHC I negativity in the proximal anagen hair bulb. This disruption of immune privilege exposes autoantigens from melanogenesis-related proteins and triggers an autoimmune response consisting of 2 phases. First, CD8+ T cells recognize melanogenesis-related proteins abnormally presented by MHC I molecules and initiate the disease on melanocytes and keratinocytes. Second, CD4+ T cells and antigen presenting cells mount an attack against MHC class II presented autoantigens that were exposed by damaged melanocytes and keratinocytes. This second response causes most of the follicular damage.[10]

AA is thought to be a multifactorial disease, involving elements of autoimmunity, genetics, and several other factors. Acute episodes of the disease are more likely to occur in individuals undergoing times of profound stress, grief, or fear, implying that certain environmental factors contribute to the pathogenesis.[11] Infectious pathogens including bacteria and viruses have been implicated in the development of other autoimmune diseases, such as diabetes mellitus and systemic lupus erythematosus, and several theories hold that AA is also the result of an infectious process. It has been postulated that AA results when individuals with a CD8+ T-cell deficiency are infected with Epstein-Barr virus, leading to the production of autoreactive T cells and the formation of ectopic lymphoid follicles.[12] Cytomegalovirus has also been theorized to play a role, after researchers[13] discovered the presence of cytomegalovirus DNA sequences in scalp biopsies taken from patients with AA. One report[14] of 12 patients who self-reported a history of mononucleosis < 6 months before the onset of AA corroborates this theory. The seasonal pattern of AA flares has been described, with relapses mostly occurring in February and March. The relative increase in relapses during these months could possibly be due to the seasonal increase in viral infections.[15]

AA occurs in genetically susceptible individuals, and several genetic markers have been identified that are associated with the development of the disease. Genome-wide association studies,[11] which uses single-nucleotide polymorphism markers to identify regions of linkage disequilibrium, were performed on 1054 unrelated AA patients and 3278 controls. Several AA-associated single-nucleotide polymorphisms were identified, clustered in eight regions across the genome and involving genes implicated in the immune system, as well as genes unique to the hair follicle.[11] In particular, the MHC antigens HLA-DQ3, DRB1*0401, and DQB1*0301 have been shown to be associated with severity of disease. Polymorphisms of interleukin 1 receptor antagonist are associated with severe early-onset of AA.[34]

The development of topical sensitizing agents

Topical sensitizers were first reported as a treatment for alopecia areata in 1978,[2] and it was found that eliciting an allergic contact dermatitis using dinitrochlorobenzene stimulated hair growth. This agent was later found to be mutagenic in 1980, by the Ames test and genotoxic by sister chromatid exchange in human fibroblasts.[16,17]

Other topical sensitizers have since replaced dinitrochlorobenzene including diphenylcyclopropenone (DPCP) and SADBE. SADBE is associated with fewer side effects than DPCP, and its absence of mutagenicity by Ames testing is more reassuring than that of DPCP, which has been found to sometimes contain a mutagenic precursor. SADBE is an ideal sensitizing agent, as it is a potent topical sensitizer, is not widely found in the environment, does not cause significant adverse effects, and does not cross-react with other chemicals.[18] Of note, SADBE is not as stable in acetone, requires refrigeration, and is more expensive compared with DPCP. Other sensitizers, including urushiol, are known to interrupt the pathologic inflammation of AA, but, due to the uncontrolled reactions, the two previously-mentioned sensitizers are preferred.

The mechanism of topical sensitizers

The mechanism of action of topical sensitizers, including SADBE, is not completely understood aside from an immune modulation from an allergic contact dermatitis. One theory describes the sensitizing compound as a hapten that binds protein substrates to create a complete antigen, which is the true immune diversion.[19] It has been reported that administration of a topical sensitizer decreases the peribulbar CD4+/CD8+ lymphocytic ratio from 4:1 to 1:1[3] and reduces the number of intrabulbar CD8+ lymphocytes and Langerhans cells.[20] The abnormal expression of class I and II major histocompatibility complex molecules on the proximal follicular epithelium that is implemented in the auto-aggressive response by CD8+ lymphocytes is down-regulated after treatment with a