

METHOD FOR TREATING EPIDERMAL OR DERMAL CONDITIONS

This application is a continuation-in-part of U.S. provisional patent application Ser. No. 60/037,098, filed Feb. 4, 1997.

This invention was made with government support from the National Institutes of Health. Accordingly, the government has certain rights in the invention.

BACKGROUND OF THE INVENTION

The invention relates to modulation of nitric oxide (NO) to treat epidermal and dermal conditions.

Organic nitrates and their gaseous metabolic end-product, nitric oxide (NO), have been implicated to date in a vast array of biologically diverse activities (Snyder et al. (1992) *Sci Amer* 5:68-77). The growth in interest in the biological effects of NO began in 1980 when it was noticed that relaxation of blood vessels (vasodilatation) no longer occurred when the endothelial layer was stripped from the vessels. The molecule mediating this effect was termed endothelial-derived-relaxing-factor (EDRF). In 1987, EDRF was shown to be nitric oxide (NO).

Three nitric oxide synthase (NOS) isoforms have been characterized. A constitutive form is found in neuronal cells (nNOS) (Schmidt et al. (1991) *Biochem Biophys Res Comm* 181:1372-77), an inducible form (iNOS) is found in macrophages (Xie et al. (1992) *Science* 256:225-28; Lyons et al. (1992) *J Bio Chem* 267:6370-74), while another constitutive form is produced by endothelial cells (eNOS) (Janssens et al. (1992) *J Biol Chem* 267:14519-22). These are also known as Types I, II and III, respectively (Pollock et al. (1991) *Proc Natl Acad Sci USA* 88:10480-84).

The role of NO in the vascular system has been shown to be extensive (Vane et al. (1990) *New Eng J Med* 323:27-36). NO participates in the regulation of systemic blood pressure as evidenced by hypertension in mice in which the eNOS gene has been knocked out by homologous recombination (Huang et al. (1995) *Nature* 377:239-42). Decreased responsiveness to NO in the pulmonary vasculature contributes to pulmonary hypertension while the vasodilator effects of NO are necessary for penile erection (Saenz et al. (1989) *New Eng J Med* 320:1025-30). Cutaneous vasculature has received some attention because the dermis has an extensive capillary network and these capillaries serve as a good model to study microcirculation in man. As in other blood vessels, the endothelium lining the dermal capillaries expresses eNOS. It has been observed that in the presence of NO, blood flow in the human skin microcirculation is remarkably increased and in the presence of inhibitors of NOS, vasodilatation is impaired (Warren, JB (1994) *EASEB J* 8:247-51; Ralevic et al. (1992) *Br J Pharmacol* 106:650-655).

Large amounts of NO are produced when macrophages are cultured with interferon-gamma (IFN- γ), tumor necrosis factor-alpha (TNF- α) or low doses of lipopolysaccharide (LPS) (Stuehr et al. (1985) *Proc Natl Acad Sci USA* 82:7738-42). Production of NO by macrophages is toxic to bacteria and parasites (Liew et al. (1991) *Immunol Today* 12(3):A17-A21). For example, resistance of mice to *Leishmania major* infection correlates with the induction of NOS in macrophages (Liew et al. (1990) *J Immunol* 144:4794-97). In mice where the iNOS gene has been knocked out, leishmania infection is severe.

The fact that NO is a neuronal messenger was first appreciated when it was shown that cerebellar granule cells

release NO after exposure to glutamate agonists (Garthwaite, J. (1991) *Trends Neurosci* 14:60-67). NOS containing neurons are found throughout the central and peripheral nervous systems (Bredt et al. (1992) *Neuron* 8:3-11). NO plays a key role in nervous system morphogenesis and synaptic plasticity.

SUMMARY OF THE INVENTION

In one aspect, the invention features, a method of treating a subject, e.g., a human, for an unwanted epidermal or dermal condition. The method includes administering to the subject, a treatment which modulates the level of nitric oxide (NO) in the skin. Conditions characterized by unwanted cells, e.g., melanocytes or keratinocytes, the proliferation of such cells, or a deficiency in apoptosis of such cells, or unwanted pigmentation, are treated by increasing the level of NO in the skin. Conditions characterized by the lack of or by an insufficient number of dermal or epidermal cells, e.g., melanocytes or keratinocytes, or a lack of pigmentation, are treated by reducing the level of NO in the skin.

In preferred embodiments the epidermal or dermal condition is: a melanocyte-related disorder; a disorder characterized by a lack of skin or hair pigmentation, e.g., graying or other loss of pigmentation of the hair; a disorder characterized by unwanted or excess skin or hair pigmentation; a disorder characterized by a deficiency in the number or activity of melanocytes; a disorder characterized by unwanted melanocyte cell death; a disorder characterized by unwanted melanocyte apoptosis.

In preferred embodiments the condition is: vitiligo; post-inflammatory hypopigmentation; post-inflammatory hyperpigmentation; or idiopathic guttate hypomelanosis (IGH).

In preferred embodiments the epidermal or dermal condition is: a keratinocyte-related disorder; a disorder characterized by deficiency in the number or activity of keratinocyte; a disorder characterized by unwanted keratinocyte cell death; a disorder characterized by unwanted keratinocyte apoptosis.

In preferred embodiments the condition is characterized by unwanted keratinocyte proliferation, caused, e.g., by a deficiency in keratinocyte apoptosis.

In preferred embodiments the condition is: an inflammatory skin disorder, e.g., eczema or psoriasis; or toxic epidermal necrolysis (TEN).

In preferred embodiments the condition is lichen planus.

In preferred embodiments the condition is sunburn.

In preferred embodiments the condition is graft versus host disease (GvHD).

In other preferred embodiments: the treatment can be administered topically or intravenously. Preferably, the treatment is repeated, e.g., it is repeated at least 1, 2, 3, 4, or 5 times.

In preferred embodiments the treatment includes: the administration of a compound; the administration of a compound which inhibits the level of NO in the skin of the subject, e.g., an inhibitor of NO synthase or an NO scavenger, e.g., a heme compound, e.g., hemoglobin, thereby preferably decreasing the level of cell death (or increasing the number of viable) skin cells, e.g., melanocytes or keratinocytes.

In other preferred embodiments the treatment includes: the administration of a compound, e.g., the administration of a compound which increases the level of NO in the skin of the subject, e.g., an NO donor compound, e.g., sodium nitroprusside (SNP) or a derivative thereof, thereby prefer-