



## Commentary

# Connexin hemichannel inhibition improves skin pathology in Clouston syndrome mice

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Mutations in the genes encoding human connexins cause a variety of clinical disorders. The vast majority of these genetic diseases cannot be cured, and current treatment aims at providing symptomatic relief, when possible. In this issue, Kuang and colleagues [1] show that administration of a monoclonal antibody raised against a connexin extracellular domain to mutant mice that expressed pathological Cx30 hemichannels associated with a Clouston syndrome mutation successfully reversed skin-related pathology. People with mutations in five different connexin genes suffer from a broad and overlapping clinical spectrum of incurable epidermal diseases, with very limited options for palliation [2]. The findings of Kuang et al. [1] represent the first mechanistic based therapy for any of the connexinopathies, as well as the first documented use of an engineered antibody to inhibit the pathological function of a mutated channel *in vivo*.

Connexins were initially identified as the protein subunits that make up the intercellular channels present in gap junctions, and subsequently found to be able to form functional hemichannels in unopposed cell membranes [3]. There are many known inhibitors of connexin channels, and a tremendous interest in translating knowledge of their action into clinical therapies [4,5]. In the case of the connexin skin diseases, functional characterization of a large number of mutations has established dysregulated hemichannel activity as the most common disease causing mechanism, reinforcing the notion that hemichannel inhibitor strategies could have therapeutic value [2,6].

In support of this idea, Kuang et al. [1] used an engineered monoclonal antibody that had been previously shown to block connexin hemichannels *in vitro*, including those formed by mutated connexins linked to human skin disease [7]. Two weeks of topical or systemic antibody treatment was sufficient to repress hyperproliferation in skin

and reduce hypertrophic sebaceous glands to normal levels in a knock-in mouse model where wild type Cx30 was replaced with the Cx30-A88V mutation that causes Clouston syndrome and displays increased hemichannel activity *in vitro* [8,9]. They further documented that in primary keratinocytes, or HaCaT cells, the antibody reduced Ca<sup>2+</sup> influx and ATP efflux through Cx30-A88V hemichannels. These findings have clinical relevance beyond Clouston syndrome, especially for the severe forms of skin disease caused by connexin mutations.

For example, Keratitis-Ichthyosis-Deafness (KID) syndrome is caused by mutations in the *GJB2* gene encoding Cx26, and all patients carrying the Cx26-G45E mutation develop recurrent cutaneous infections, septicemia, and die during infancy [2,6]. In these lethal cases, the conventional clinical trial process is not practical due to the scarcity of patients and their medical urgency at presentation. The positive results obtained by Kuang et al. [1] in treatment of a mouse model of Clouston syndrome may offer clinicians treating these KID patients with poor prognoses a therapeutic alternative. The same monoclonal antibody used in this study was previously shown to inhibit hemichannels formed by the Cx26-G45E mutation *in vitro* [7]. Thus, the inhibitory antibody approach outlined by this study could offer a new treatment option in severe cases of KID syndrome, and help children born with the Cx26-G45E mutation survive infancy.

Hemichannel dysfunction is emerging as a general pathological mechanism in several of the connexinopathies. This idea was recently reinforced by the demonstration that a truncated mutation of Cx32 (Cx32-R220X) which causes the X-linked form of Charcot-Marie-Tooth disease, displayed severely impaired hemichannel gating. In this case, the mutant protein retained the ability to form functional intercellular channels (reviewed in [3]), whereas Cx32-R220X hemichannels could not open in response to physiologically induced elevations of cytosolic Ca<sup>2+</sup>. In this case, the proposed intervention aims at restoring proper hemichannel activity with the extracellular application of a peptide resembling the C-terminal cytoplasmic domain of Cx32, further providing a proof-of-concept that hemichannels are potent druggable targets to treat human diseases [10].

Currently, there are only palliative treatments available for the connexin-mediated channelopathies. The elucidation of the mechanisms whereby connexin mutations alter hemichannel activity and result in epidermal pathology, coupled with the development of strategies to disrupt those mechanisms has lead us to the point

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where their efficacy can now be tested in mouse models that replicate human disease. As demonstrated by Kuang et al.[1], blocking aberrant connexin hemichannel activity with specific monoclonal antibodies offers a new treatment paradigm for some of the incurable human pathologies linked to connexin mutations. It also provides a valuable new tool to probe why increased hemichannel activity is so disruptive to the epidermis, knowledge that could in turn lead to additional novel therapeutic approaches to treating connexin disorders of the skin and beyond.

### Declaration of Competing Interest

The authors declare no conflicts of interest.

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