## *In vivo* and *in vitro* testing with PEGylated nanoparticles

*To the Editor:*

I read with interest the article by Troelnikov et al[1] describing 3 patients with a history of prior reactions to polyethylene glycol (PEG)-containing materials, with all 3 of them demonstrating positive skin test reactions to an mRNA vaccine containing PEGylated nanoparticles but 2 of the 3 exhibiting negative prick and intradermal skin test responses to PEG and other PEG-containing materials. These skin test results were supported by positive basophil activation test results in response to PEGylated nanoparticles but not to PEG alone.

The article raises a number of questions. Why would these patients who "had past positive PEG skin testing" have lost this reactivity? If the patients react only to PEGylated nanoparticles, how would this explain their prior reactions to other forms of PEG?

As the authors state, these results suggest that PEGylated nanoparticles may be better able to cross-link cell-bound IgE or stimulate complement activation–related pseudoallergy reactions. A serum-specific IgE assay to PEGylated nanoparticles could help answer the question of an IgE-mediated versus complement activation–related pseudoallergy reaction.

Although the patients had positive vaccine skin test results, whether they would react to actual vaccine administration by either single or graded dosing is unknown. It will be important to perform the vaccine basophil activation test on patients who have actually had apparent allergic reactions to vaccine administration to support the notion that such reactions are caused by the PEGylated nanoparticles, because to date, no cases of vaccine anaphylaxis have been definitively linked to PEG.

*John M. Kelso, MD*

From the Division of Allergy, Asthma and Immunology, Scripps Clinic, San Diego, Calif. E-mail: kelso.john@scrippshealth.org.

Disclosure of potential conflict of interest: The author declares that he has no relevant conflicts of interest.

### REFERENCE

1. Troelnikov A, Perkins G, Yuson C, Ahamdie A, Balouch S, Hurtado PR, et al. Basophil reactivity to BNT162b2 is mediated by PEGylated lipid nanoparticles in PEG allergic patients. J Allergy Clin Immunol 2021;148:91-5.

Available online June 26, 2021.
https://doi.org/10.1016/j.jaci.2021.06.004

## Reply

*To the Editor:*

We thank Dr Kelso for his correspondence[1] regarding our recent article[2] in which we demonstrated that polyethylene glycol (PEG)-containing substances are not reliable surrogates for skin testing for allergy to the BNT162b2 COVID-19 vaccine. Although PEGylated liposomal drugs induced basophil activation *ex vivo* in 3 patients with PEG allergy, PEG alone did not. Dr Kelso highlights 3 important questions raised by this research that are worth discussing.

The first question relates to why 2 patients who had previously demonstrated a positive skin test result in response to PEG-containing substances appeared to have lost their reactivity. We were also intrigued by this; however, loss of skin test positivity is not uncommon in drug allergy,[3] and has recently been documented in patients with PEG allergy.[4] As these 2 participants reacted to PEGylated liposomal vaccine, we suggest that the inconsistent skin testing results are a consequence of PEG being poorly allergenic in its native state. In support of this, we present a fourth patient, a 38-year-old female with a history of anaphylaxis following PEG3350-containing bowel prep and PEG excipient–containing drugs. Her skin prick testing was initially performed at another institution, and the results were negative in relation to PEG200, 400, 600, 3350, 2000, and 6000. However, upon oral challenge with 35 mg of PEG3350 (Movicol; Norgine, Sydney, Australia), definite urticaria was observed at multiple sites within 30 minutes. Six weeks later, the patient underwent skin testing panel as we described elsewhere[2] with a positive result in response to the BNT162b2 vaccine. In agreement with our original case series, basophil activation testing demonstrated positivity in response to the vaccine and to PEGylated liposomal doxorubicin (Fig 1) but not to PEG alone. This case suggests that skin testing with "free" PEG does not necessarily equate to hypersensitivity, and as such, skin testing with PEGylated liposomal drugs is useful not only for the assessment of vaccine risk but also in the diagnosis of PEG allergy.

The second question relates to the mechanism by which PEGylated liposomes induce basophil activation. Dr Kelso suggests measuring vaccine-specific IgE level in these patients as an indicator that basophil activation in response to the vaccine is IgE dependent as opposed to involving IgE-independent mechanisms such as complement activation–related pseudoallergy. Although this is valid, there are technical issues with measuring antibodies against PEG or against conformationally intact liposomes.[5] The involvement of IgE can, however, be directly tested by selective depletion of IgE in an indirect basophil activation test. This is something that we intend to address.

The third question is whether these patients are representative of those experiencing anaphylaxis following administration of the vaccine. We certainly believe that these patients are at risk given their history of anaphylaxis in response to PEG and their *in vitro* and *in vivo* reactivity to the vaccine, and we therefore agree with the existing recommendations against using PEGylated liposomal vaccines for vaccination of patients with PEG allergy. Nonetheless, we agree that it is important to now apply basophil activation testing in those who react to administration of the vaccine, and we invite contribution of patients from outside our small local population in South Australia.

*Griffith Perkins, BSc (Adv)*[a,b]
*Alexander Troelnikov, MBBS, BMedSci (Hons)*[a]
*Pravin Hissaria, MD, MBBS, FRACP, FRCPA*[c]

From [a]SA Pathology, Adelaide, Australia; [b]the School of Biological Sciences, University of Adelaide, Adelaide, Australia; and [c]the Immunology Department, Royal Adelaide Hospital, Adelaide, Australia. E-mail: pravin.hissaria@sa.gov.au.

Supported by the Hospital Services Charitable Gifts Board, Royal Adelaide Hospital.
Disclosure of potential conflict of interest: The authors declare that they have no relevant conflicts of interest.