

Standardization of allergenic preparations

Comments with reference to the second edition of the common Nordic guidelines for registration of allergenic preparations

A. BACKMAN, L. BELIN, S. DREBORG, R. HALVORSEN, H.-J. MALLING & B. WEEKE*

Twelve years ago, the Committee for Allergen Standardization within the Nordic Association of Allergology first proposed a system for standardization of allergenic extracts based on careful selection of raw materials, characterization by CIE-CRIE methods, biological standardization of in-house standards, and RAST inhibition for control of the inter-batch variation in potency. Later the Nordic Council on Medicine recommended that this system should be used for documentation of allergenic preparations intended for registration in the Nordic countries.

Since that time new knowledge has accumulated. The sub-committee for allergen standardization within the International Union of Immunological Societies has established International Standard preparations (IS) of seven common inhalant allergens, which are now available to national authorities, manufacturers and scientists. They should be used as yardsticks for national standards and in-house standards produced by national agencies, allergen manufacturers and research laboratories. The composition of the IS preparations is well established, but their potency is arbitrary. For each standard, the content of an ampoule of a standard is given the potency 100 000 International Units (IU), i.e. 100 000 IU/ml when reconstituted with 1 ml of diluent. The excellent work

performed by many scientists to establish these standards has produced much new information.

Many manufacturers were opposed to the first edition of the Nordic guidelines published in 1982 for registration of allergenic preparations. One conflicting issue was the requirement for toxicity studies. In the new edition of the Nordic guidelines, which recently appeared*, this issue has been clarified. Furthermore, the chapter on clinical documentation of allergenic preparations has been extended and the difference between documentation of allergenic preparations intended for diagnostic and therapeutic use has been pointed out.

Another controversial part of the first edition was biological standardization. The aim of biological standardization was to establish a common biological unit for different types of allergens. A critical point for meaningful biological standardization is that patients included in the standardization procedure are consecutive adult patients with inhalant allergy, clinically sensitive to the allergen in question, attending a university allergy outpatient clinic. With these criteria for inclusion of test subjects, biological standardization in different parts of Europe have yielded similar results,

* Members of the "Committee on Allergen Standardization" within the Nordic Association of Allergology and Clinical Immunology.

i.e. the median sensitive patient in samples (groups) of 20 test subjects, showed similar skin sensitivity.

In the Nordic guidelines, the authorities prescribed biological standardization of both diagnostic and therapeutic allergenic preparations. In the second edition they provide a method for biological standardization by quantitative skin prick testing, to be complied with by manufacturers. However, it is indicated that other skin prick test methods shown to correlate with the method prescribed may also be used. Results of a simple method for biological standardization, including only 4-8 skin prick tests on the forearm of each patient, has been shown to correlate well with those obtained with the original method. Similarly, all other methods suggested in the Nordic guidelines may be replaced by other methods, provided documentation is shown that results obtained with the methods chosen correlate with and give the same information as the proposed method.

It will be difficult to select patients allergic to food allergens or to laboratory animals, who fulfill the inclusion criteria for biological standardization. Patients allergic to foods and small furred animals do not show the same type of symptoms, the same age distribution (adults), are not similarly exposed, and their sensitization does not show the same time course as that of patients with respiratory allergy. The commonest group of patients sensitive to food allergens is small children, who often exhibit severe eczema, gastro-intestinal symptoms and sometimes also respiratory symptoms during a short period of their lives and then come to tolerate the allergen in question. Patients allergic to small furred animals are either children who keep them as pets, or adults with occupational allergy. For patients with allergy to small furred animals it is usually easy to avoid further contact as they are not as common as dogs, cats, and mites.

Thus, it is difficult to find test subjects for biological standardization of allergenic preparations based on foods, small furred animals, and many other less common allergen sources. We propose that only common airborne allergens should be standardized in full accordance with the Nordic concept. Any allergens that differ from common inhalant allergens as regards type of exposure, sensitization pattern, patient's clinical picture and age at onset, should be exempted from biological standardization. In Table 1 we suggest five allergen categories which should be available for clinical use. It would be a problem if important allergens were no longer available to clinicians because of too rigorous demands for biological standardization which are impossible for laboratories and manufacturers to meet. As knowledge about alimentary and less common airborne allergens increases, upgrading of lower category allergens on the proposed category ladder, should be encouraged.

Only class A should be available for immunotherapy. We regard the present method for biological standardization as unfeasible for standardization of allergens belonging to groups B, C and E. Potency calibration by RAST inhibition or similar techniques is suggested when sera from several sensitized individuals with specific IgE antibodies to the allergen in question are available. As an alternative, immunoblotting, that requires less serum than repeated RAST inhibition experiments, can be used as a means of semi-quantitative estimation of the relative potency of new batches. Therefore, these should be the preferred techniques for uncommon allergens which are of clinical importance but difficult to standardize.

Category D, (special extracts), are extracts produced by research laboratories or by manufacturers for scientific purposes. To continue research regarding the possible role of less well-known allergens in the environment, extracts of such allergens should be exempted from registration and allowed to be produced, shipped and used for diagnostic tests. Finally, at the discretion of the responsible clinician, fresh fruits, meat, vegetables, etc., may be used for skin prick test according to the prick-prick

* Nordic Council on Medicine, Registration of Allergenic Preparations, Common Nordic Guidelines, NLN Publication No. 23, Uppsala 1989.

Address: Box 607, S-751 25 Uppsala, Sweden.

method for diagnosis of sensitization to food allergens.

In vivo testing with biologically standardized allergenic preparations, provides information of great importance to clinicians.

It has been discussed whether biological standardization is a relevant method for standardization of allergenic preparations for therapeutic use. The Nordic guidelines prescribe that also therapeutic allergenic preparations should be labelled in BU/ml. Whenever possible, the potency also should be given in International Units (IU/ml). Some investigators prefer allergen preparations intended for immunotherapy to be standardized with regard to major allergen content. Recent reports indicate a correlation between the content of the dominating major allergen in preparations of common inhalant allergens and their biological

activity, i.e. 1 BU corresponds to approximately 1 ng ($\pm 50\%$) of major allergen. Therefore, in the future, determination of major allergen content might be a suitable method for standardization of allergen preparations intended for immunotherapy, i.e. category A, and it would soon be possible to use this standardization method for categories B, C and D preparations. However, based on present knowledge, we do not find it relevant to replace biological standardization of category A diagnostic extracts with major allergen determination.

It has also been proposed to replace biological units with units related to the effect of immunotherapy, e.g. a change in skin sensitivity. Such a unit might be of value for standardization of modified allergens, since these allergens cannot be standardized on the basis of their allergenic potency. However, as only limited documenta-

Table 1.

Proposed types of allergens for human use, with examples. Only category A allergen preparations may be used for immunotherapy, provided that they are in accordance with the Nordic Guidelines. Allergens made available to clinicians should be stable, i.e. there should be an in-house freeze-dried standard. Commercial products intended for diagnostic use should be marketed freeze-dried or dissolved in glycerol diluent.

Type of allergen	Characterization*	Calibration**	Biological standardization
A Common inhalant allergens	+	+	+
Common insect venoms	+	-	$\mu\text{g venom}$
B Common foods	+	+	-
cow's milk			
hen's egg			
fish			
soy			
Small furred animals	+	+	-
mouse			
rats			
guinea pigs			
hamsters			
Insects	+	+	-
cockroaches			
storage mites			
C Less common foods	+	-	-
Uncommon animals, insects etc.	+	-	-
D New allergens under study	+/-	+/-	+/-
E Fresh material for prick-prick testing	-	-	-

* Characterization means that the composition is studied by biochemical methods, i.e. protein assays, determination of carbohydrate content, etc., and immunochemical methods revealing the allergenic content, i.e. CIE/CRIE and related techniques or immunoblotting.

** Calibration means that the potency is determined in relation to a standard, e.g. an in-house standard of an allergen manufacturer or an IS (and adjusted to a suitable strength).

tion is available, this concept must be regarded as a proposal for the future, as was biological standardization 12 years ago.

In summary, the revised Nordic guidelines are a marked step forward. In combination with the International Standards set up by IUIS and WHO, most common inhalant allergens can be standardized according to the Nordic guidelines; less common allergens used only for diagnosis (skin prick test) should be exempted. At the same time, manufacturers should be encouraged to increase their efforts to standardize these less common allergens; we propose standardization by molar weight. We

also propose new methods which would allow standardization of modified allergenic preparations as well. Future activities must be concentrated on equalizing regulatory control throughout Europe.

Address:

Ass. Professor *Sten Dreborg*
Department of Pediatrics
Faculty of Health Sciences
University Hospital
S-581 85 Linköping
Sweden

This document is a scanned copy of a printed document. No warranty is given about the accuracy of the copy. Users should refer to the original published version of the material.