

Novel Nanofluidic IgE Assay versus a Reference Method: A Real-World Comparison

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Keywords

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Abstract

Background: Clinically meaningful specific IgE determination is an important step in the diagnosis of allergic diseases. While patient's history and skin prick tests are available during the medical visit, most IgE immunoassays require hours to several days to be available. Recent developments in the field of nanofluidic technology open new horizons for point-of-care management of this unmet medical need. **Objective:** This study aimed to compare IgE diagnostic agreement between a nanofluidic assay (abioSCOPE[®]) and a laboratory reference method (Phadia Laboratory System[®]) in a real-world clinical setting. **Methods:** Sera from 105 patients whose routine allergy diagnostic workup required a blood sampling were used to compare the novel nanofluidic IgE assay to a reference method in a blind manner for a panel of five respiratory allergens. To assess the agreement between methods, patient records were reviewed by four independent experts to establish the final diagnosis. Experts were

blinded to the IgE serological method used, but had access to patient history, skin prick tests, and blood test results. **Results:** Analytic agreement between the two methods was 81% for the tested panel of allergens (ranging from 77 to 89%). The overall agreement in clinical diagnosis decision taken by the expert panel was 94.6% with the nanofluidic IgE assay when compared to the reference method. **Conclusion:** The nanofluidic IgE assay, as determined through an evaluation based on clinical history, skin prick tests, and IgE measurement, is a valuable tool for allergy experts to identify patients' sensitization patterns at the point of care, and for routine IgE diagnostic workup.

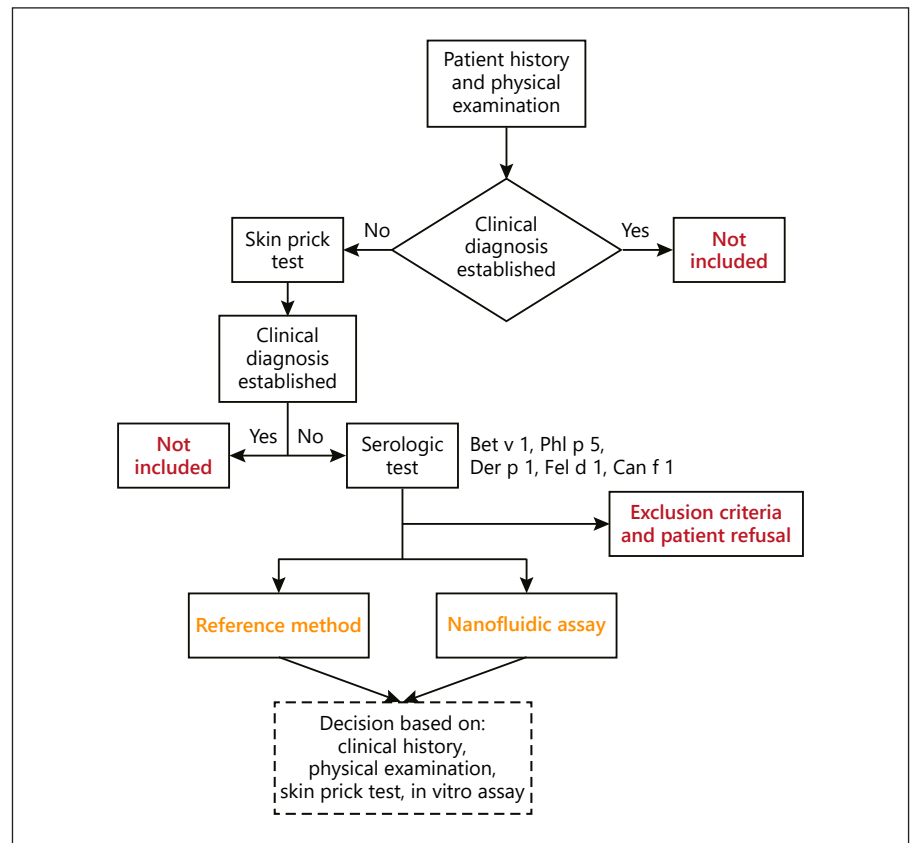
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Introduction

Diagnosis of allergy is complex and relies on the integration of information coming mostly from patient history, results of in vivo skin testing, and serological assays.

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Fig. 1. Study design. Patients visiting the CHUV Allergy and Immunology Division (Lausanne, Switzerland) between June 2013 to April 2014 and May 2015 to November 2016 and whose routine diagnostic workup required skin prick test and a blood sampling were enrolled to compare nanofluidic assay and reference method in a blind manner for a panel of 5 immunoassays: Bet v 1, Phl p 5, Der p 1, Fel d 1, and Can f 1. In order to assess the agreement in clinical diagnosis, patient records were reviewed by 4 independent allergy experts to establish final diagnosis.



IgE antibody assays were historically performed with whole allergen extracts immobilized on a solid phase and incubated with patient's serum or plasma [1]. Today's advances in molecular biology, protein production, and structural biology are allowing the development and access to IgE immunoassays with allergens purified from either their natural sources or a heterologous expression system. Under the term "molecular allergology" hides a revolutionary concept in which sensitization to single allergenic components are identified [2]. This offers several advantages over extract-based assays, such as the identification of a patient's genuine sensitization pattern allowing for a risk evaluation of severe reactions, improved interpretation of cross-reactivity, and better indications for specific immunotherapy [3]. Molecular allergology is today considered an important complementary approach to in vivo skin testing, as illustrated by the European Academy of Allergy and Clinical Immunology's (EAACI) recently endorsed user's guide presenting this concept [2].

Nevertheless, where clinical history and results from in vivo skin testing can be obtained within a unique medical consultation, IgE antibody assays are most often outsourced to medical laboratories and performed on large

automated equipment. This long turnaround time delays treatment initiation, affects patient motivation, and increases the number of patient visits to the treating clinicians. Key requirements for a point-of-care platform meeting physicians' needs include compatibility with capillary whole blood measurement to avoid time-consuming processing, short turnaround time (typically below 20 min), easily interpretable, objective, and accurate results, and a handling that necessitates as few steps as possible to mitigate the risk of analytical errors.

Recent developments in the field of nanofluidic technology open new horizons for point-of-care testing. The nanofluidic principle relies on the specimen passing through a channel of nanometric dimensions in which molecular interactions are strongly enhanced [4]. In this channel, molecules, typically allergens, are specifically immobilized to the solid phase and are able to capture allergen-specific IgE passing through. This novel and quick technology therefore brings serological assays directly to the physician's office, assays that are today mostly outsourced to the local laboratory.

In this study, we evaluated for the first time the diagnostic efficacy of a nanofluidic assay (NFA) (abio-

SCOPE[®], Epalinges, Switzerland) as an aid in the diagnosis of allergic diseases. We found a good agreement between test results obtained with the NFA as compared to the reference method (RM) (Phadia Laboratory System, formally named ImmunoCAP[®], ThermoFisher Scientific, Uppsala, Sweden), considered as a quasi-gold standard in IgE antibody assays. The analytic agreement between the two methods was 81% for the tested panel of allergens (ranging from 77 to 89%). Moreover, we found that the overall agreement in final clinical diagnoses taken by a panel of allergy experts who had access to patient history, skin prick tests (SPTs), and IgE serological assay results, but were blinded to the IgE antibody method used, was 94.6% with the NFA when compared to the RM. Taken together, these data strongly support the NFA as a valuable tool to identify allergen sensitization at the point of care and to support allergy specialists in the routine diagnostic workup of IgE-mediated disorders.

Materials and Methods

Clinical Study Design and Expert Committee

The study was designed as a single-center, laboratory blind phase I clinical trial. One hundred and five adult patients, aged 18–65 years, referred at the CHUV Division of Immunology and Allergy (Lausanne, Switzerland) between June 2013 to April 2014, and May 2015 to November 2016, were enrolled into the study (Fig. 1). This panel of patients was constituted of patients for whom the clinician in charge considered it necessary to proceed to IgE blood tests because of an uncertain diagnosis despite a previous workup of SPTs. Sera were first tested for the specific questions raised by the clinician, then aliquoted and stored at –80 °C at the central laboratory, CHUV, Lausanne until tested for the panel of IgE assays in the current study (Fig. 1). Exclusion criteria were as follows: positive HIV viremia in the 12 months preceding the study, use of any investigational or non-registered product (drug or vaccine), chronic administration of immunosuppressive or immunomodulatory drugs within the 6 months preceding the study, administration of any immunoglobulins, immunotherapy, or blood products in the 3 months preceding the study or planned during the study period, participation in another experimental protocol, any health condition that could interfere with the evaluation of the study endpoint in the opinion of the investigator, personal or family history of congenital or hereditary immunodeficiency, use of antihistamine, systemic steroids, cytokines, biologicals, or chemotherapeutic drugs in the week preceding the skin test evaluation. Informed consent was obtained from all subjects before participating to the study. The study protocol was approved by the Institutional Review Board from Lausanne University Hospital (IRB No. 309/12 + amendment) prior to the initiation of the study and was in line with the declaration of Helsinki in its latest revision [5] and registered in www.clinicaltrials.gov under reference NCT03599908.

The primary endpoint of the study aimed to assess the agreement in the final clinical diagnosis based on IgE results according

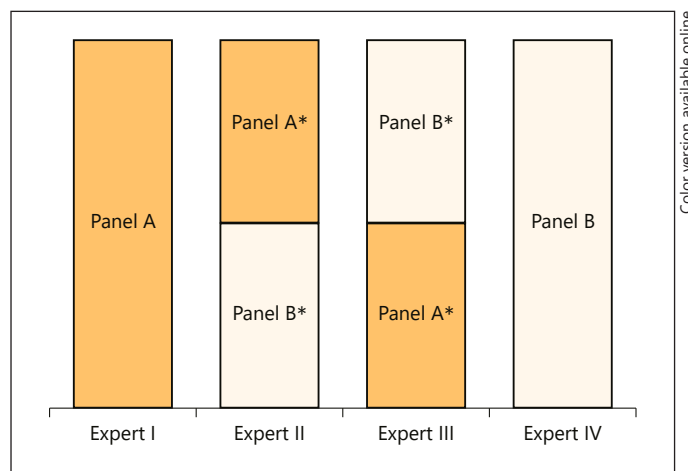


Fig. 2. Impact study patient distribution. Patients were randomly distributed into two panels (A and B), according to the IgE assay technique (reference method vs. nanofluidic assay). For experts II and III, panels were distributed alphabetically.

to assay methods, i.e., sIgE by NFA versus RM (Fig. 2). To do so, patient records were reviewed by an expert panel of four independent allergologists blinded to the serological methods used. Patients ($n = 105$) were each distributed randomly into 2 panels (A [$n = 105$] and B [$n = 105$]) according to the IgE assay technique used (NFA vs. RM). Each of the four experts were individually assigned to either Panel A, first half ($n = 53$) of Panel A and second part of Panel B ($n = 52$), first half of Panel B and second part of Panel A, and Panel B, respectively.

Based on the experts' decision, blind investigators computed the agreement in a final clinical diagnosis. In a case of non-concordance between two experts judging a clinical situation based on the same IgE methodology, the final diagnosis was based on the decision of a fifth, blind expert.

Skin Prick Tests

SPTs were performed using commercially available allergen extracts for 20 common allergens (Allergopharma [Germany], Stallergenes [France]). Tests were carried out and interpreted according to the established criteria recommended by EAACI guidelines [6], by using standardized 1-mm single-pick polymethacrylate points (Stallergenes). Sodium chloride (0.9%) and histamine hydrochloride (10 mg/mL) served as negative and positive controls, respectively. Wheal and flare reactions were measured after 15 min and were considered positive with a wheal diameter ≥ 3 mm.

IgE Antibody Assays

All patients' sera were analyzed for allergen-specific IgE using the NFA and the RM in a blind manner for a panel of five IgE assays: Bet v 1, Phl p 5, Der p 1, Fel d 1, Can f 1. Samples already tested for a specific allergen with the RM were not re-tested.

Both quantitative IgE antibody methods used rely on the immobilization of allergenic components purified from either their natural source or from a heterologous expression system on a solid phase (Table 1). Allergens on the RM were immobilized in a three-dimensional cellulose polymer forming a sponge-like struc-

Table 1. Comparison of reference method and nanofluidic assay

	Reference method	Nanofluidic assay
<i>Similarities</i>		
Method		“Sandwich” immunoassay
Analyte measured		Allergen-specific IgE antibodies
Reported value		Quantitative (kU _A /L [IgE-specific assay] or kU/L [total IgE])
Calibration		Traceable to the WHO Third IgE reference, preparation 11/234
<i>Differences</i>		
Method	Fluorescence enzyme immunoassay	Fluorescent immunoassay
Solid phase	3-Dimensional biofunctionalized sponge	2-Dimensional biofunctionalized solid surface
Assay time	105 min for the first results	11 min for the panel of 6 tests
Sample volume	400 µL initial for a panel of 6 tests	50 µL for the 6 tests
Limit of detection	0.1 kU _A /L	0.70 kU _A /L
Assay reportable range	0.35–100 kU _A /L	0.70–100 kU _A /L
Place of use	Clinical laboratories	Point of care

Table 2. Reference method and nanofluidic assay allergens used in comparison

Allergen	Reference method	Nanofluidic assay
Bet v 1	Recombinant (t215)	Recombinant
Phl p 5	Recombinant, isoform b (g215)	Recombinant, isoform a
Der p 1	Natural (d202)	Natural
Fel d 1	Recombinant (e94)	Recombinant
Can f 1	Recombinant (e101)	Natural

ture with high allergen-binding capacity. In contrast, the allergens analyzed on the NFA were immobilized in a two-dimensional solid phase in the sensing area of the nanofluidic sensors. Venous whole blood was collected following standard venipuncture procedure. Serum was prepared from the whole blood, anonymized, and kept frozen in controlled storage areas until being analyzed on the RM, using the tests t215 (Bet v 1), g215 (Phl p 5b), d202 (Der p 1), e94 (Fel d 1), and e101 (Can f 1) (ThermoFisher Scientific, Uppsala, Sweden) or on the NFA with a capsule quantifying IgE antibodies to the allergens Bet v 1, Phl p 5a, Der p 1, Fel d 1, and Can f 1. Both test procedures were performed following the manufacturer's instructions. Both assays are quantitative and calibrated with a calibrator traceable to the WHO IgE reference standard, preparation 11/234 (National Institute for Biological Standards and Control, Potters Bar, UK). The RM is a singleplex device that reports results in the range of 0.35–100 kU_A/L, whereas the NFA is an oligoplex platform (one capsule contains the five tests of the study) reporting results in the range of 0.70–100 kU_A/L. Table 1 and 2 highlight the similarities and discrepancies between both IgE antibody assay methods.

In practice, the patient sample is mixed with a solution containing a fluorescent monoclonal anti-human IgE antibody and is then deposited into a capsule containing nanofluidic sensors, each coated with a different allergen, that are then analyzed in a fully automated fluorescent microscope, the abioSCOPE® (Abionic SA,

Epalinges, Switzerland). Thanks to its small footprint, the abioSCOPE® is located in the medical doctor's office, either on a desk or fixed to the wall.

Statistics

Demographic and baseline characteristics were analyzed by descriptive statistics. A positive result for the quantitative allergen-specific IgE tests was defined as a concentration ≥ 0.70 kU_A/L. Three different agreement percentages were computed: the positive percent agreement and negative percent agreement were calculated with the proportions of agreement for the average of their positive and negative responses. The overall percentage agreement was calculated as the ratio, expressed in percent, of the total number of results minus the number of discrepancies divided by the total number of results.

Cohen's kappa analysis for agreement of detection results was also applied, and kappa values were categorized as almost perfect (0.80–1.00), substantial (0.60–0.79), moderate (0.40–0.59), fair (0.20–0.39), and poor (below 0.20) [7]. All calculations were performed in Excel for Windows, version 1810 (build 11011.20074).

Results

A total of 105 patients were included into the study from June 2013 to November 2016. All patients for whom clinicians considered it necessary to request a complementary IgE blood test for diagnostic purposes were asked to participate in the study and had to give informed consent. There were 33 (31.4%) males and 72 (68.6%) females with a mean age of 36.3 ± 14.6 years. Fifty-three (50.5%) patients had positive SPTs for birch pollen, 61 (58.1%) for grass pollen, 45 (42.9%) for cat dander, 21 (20.0%) for dog dander, and 41 (39.0%) for *Dermatophagoides pteronyssinus*. The most frequent reason for con-

Table 3. Description of the study allergic patient population based on reference method sIgE level

	Bet v 1	Phl p 5	Der p 1	Fel d 1	Can f 1
<i>Reference method</i>					
N (between 0.70 and 100 kU _A /L)	51	34	18	38	8
Average, kU _A /L	23.4	24.5	10.6	16.6	4.4
SD	28.1	29.2	23.3	26.8	2.8
Median, kU _A /L	10.8	11.5	3.6	6.0	3.4
<i>Nanofluidic assay</i>					
N (between 0.70 and 100 kU _A /L)	29	24	20	32	20
Average, kU _A /L	20.7	27.1	9.6	21.4	49.3
SD	30	31.5	21.9	30.4	47.2
Median, kU _A /L	7.9	11.5	2.6	6.5	35

Detection range was 0.70–100 kU_A/L and the total number of patients was 105. kU_A/L, kilounits of antibody per liter; SD, standard deviation.

sultation was respiratory allergy ($n = 87$) with a primary diagnosis of allergic rhinoconjunctivitis ($n = 40$), allergic asthma ($n = 1$), or both ($n = 46$). Twenty-eight had seasonal, 16 perennial, and 43 combined seasonal and perennial symptoms. Among allergic patients, 34 had no other additional diagnoses of allergic disease, 33 patients had 2 diagnoses, 13 had 3 diagnoses, and 8 had more than 3 diagnoses. Eleven patients had no allergic disease. An additional diagnosis of food allergy was made in 41 patients, mostly due to cross-reactivities to pollens ($n = 37$) (oral allergy syndrome). Eighteen patients presented with drug allergies, 16 with non-allergic rhinitis, 12 with systemic anaphylaxis, 7 with gastroesophageal reflux disease, 4 with non-allergic asthma, 3 with *Hymenoptera* sp. allergy, 6 with chronic spontaneous urticaria, and 2 with eosinophilic esophagitis.

A low prevalence of poly-sensitizations was observed in the whole cohort, with median allergen-specific IgE values comprised between 3.4 and 11.5 kU_A/L after the RM and between 2.6 and 35 kU_A/L for corresponding tests after the NFA (Table 3).

The analytic agreement between the two IgE antibody assay methods, i.e., the laboratory RM and the point-of-care NFA, was assessed for five major respiratory allergens: Bet v 1, Phl p 5, Der p 1, Can f 1, and Fel d 1. Table 4 shows that overall percent agreement varies from 77% to 89%. The negative percent agreements were consistently higher (83–100%) than the positive percent agreements (54–97%), highlighting that different technologies and source of allergen components result in different outcomes. Table 5 shows overall agreements for SPT versus IgE antibody assays, that were comprised

Table 4. Agreement between nanofluidic assay and reference method sIgE per allergen IgE antibody assay

	<i>n</i>	P	N	NPA	PPA	OPA	Kappa
Bet v 1	105	51	54	100	57	79	0.58
Phl p 5	105	34	71	99	68	89	0.72
Der p 1	105	18	87	85	39	77	0.23
Fel d 1	105	38	67	90	66	81	0.57
Can f 1	105	8	97	83	38	79	0.11
Overall	525			90	58	81	0.51

Total number of patients was 105. Number of patients with positive results on reference method (P, corresponds to >0.70 kU_A/L) and patients with negative results on reference (N) were used as true positive and true negative. NPA, negative percent agreement; PPA, positive percent agreement; OPA, overall percent agreement.

between 67 and 90% for the RM and between 55 and 72% for the NFA.

To put these values into a real-life clinical setting, the diagnostic impact of using allergen-specific tests on the NFA versus similar tests on RM was evaluated retrospectively by two panels of allergy experts, each having access to all patient's data but were blinded to the serological method. The agreement in the clinical diagnosis decision was then computed and found to be highly concordant, ranging from 92.4 to 99.1% (Table 6). The lower agreements were found in patients sensitized to dog dander and house dust mite, for which the mean allergen-specific IgE levels were the lowest of the study cohort.

Table 5. Agreement of nanofluidic assay and reference method compared to skin prick test results

	<i>n</i>	Nanofluidic assay				Reference method			
		NPA	PPA	OPA	Kappa	NPA	PPA	OPA	Kappa
Bet v 1	96	100	49	72	0.46	95	83	89	0.85
Phl p 5	95	97	38	59	0.28	50	52	67	0.40
Der p 1	95	80	22	55	0.02	98	39	73	0.40
Fel d 1	99	87	53	71	0.41	98	78	90	0.77
Can f 1	93	85	29	72	0.14	99	29	83	0.36
Overall	478			66	0.29			80.4	0.62

NPA, negative percent agreement; PPA, positive percent agreement; OPA, overall percent agreement.

Table 6. Positive and negative percent agreements calculated in function of diagnostic made by expert panel

	<i>n</i>	P	N	D	% agreement
Cat dander	105	36	68	1	99.1
Grass pollen	105	52	50	3	97.1
Birch pollen	105	41	59	5	95.2
Dog dander	105	10	87	8	92.4
House dust mite	105	30	67	8	92.4
Overall					94.6

Expert panel had nanofluidic assay sIgE or reference method sIgE result. Number of patients for whom the expert panel agreed that the patients were sensitized (P) or not sensitized (N) to a given allergen. Diagnostic discrepancies within expert panel is represented by D. Total number of patients was 105.

Discussion/Conclusion

IgE antibody assay developments have always followed the advances in physical, material, and chemical sciences as well as micro- and nanofabrication technologies [4, 8], from paper-based assays in the original RAST to a cellulose-based polymer covalently coated with crude allergen extract or purified components, which is currently the basis of tests on the RM. More recently, microarray tests containing more than one hundred components on a stamp-size slide have reached the market, such as the ImmunoCAP ISAC® (ThermoFisher Scientific, Uppsala, Sweden) [9]. The latest development in allergy serological assays takes advantage of nanofluidic technology. Nanofluidic channels modify diffusional behavior of biomolecules and thereby reduce assay time [4]. Analytes such as IgE can be quantified in the

picomolar range in as little as five minutes. The nanochannels are assembled in a capsule that can contain up to fourteen allergy tests. Of particular interest, the nanofluidic-based sensors analyzed on the NFA provide results in minutes and demand limited sample preparation and handling time. As a consequence, the NFA is intended to be used at the point of care, allowing identification of a patient's sensitization during their medical visit.

In parallel to these technological developments, a better understanding of molecular allergology today allows for quantification of IgE levels to single allergenic components. Sensitization to defined allergens, such as food LTP, may be associated with a higher risk of severe reactions when a patient is exposed to such an allergen [10–12]. IgE thresholds have also been determined to evaluate when food reintroduction may be possible with a reasonable safety or to help in deciding whether oral food challenge may be required [13]. Cross-reactivity can be better evaluated and the genuine sensitizer from a cross-reactive allergen can be identified.

This study aimed at comparing a novel quantitative IgE antibody assay intended to be used at the point of care to the quasi-gold standard laboratory IgE antibody assays, the Phadia Laboratory System. Since the objective of this study was clearly to go beyond a crude comparison of assay performances in an *in vitro* setting where usually the sample pool is artificially enriched with high RAST classes samples, we put the emphasis on evaluating the final outcome of these techniques in real-world settings, i.e., by joining inputs from the clinical history, SPTs, and IgE assays. To the best of our knowledge, this is the first study that compares the clinical diagnostic outcome resulting from the analysis of patients' medical history and the results of SPTs and of two dif-

ferent IgE antibody assay methods. As the diagnosis of IgE-mediated diseases precisely relies on the combined interpretation of data coming from patient's history, physical examination, in vivo SPT results, and in vitro immunoassays, it is of highly relevant interest to evaluate the impact of different serological methods on the final clinical diagnosis taken in the broad context of the patient.

In order to best reflect routine practice, patients were only included if a serological assessment was deemed useful by the clinician. Meaning, most diagnoses were reliably based on patient history, clinical examination, and skin tests and were not included in this study. This introduces a selection bias towards a difficult-to-diagnose population. It also explains the low median value (IVD capsule aeroallergen 2.6–11.5 kU_A/L; ImmunoCAP 3.4–11.5 kU_A/L) of IgE measurements in both methods. Nonetheless, overall percent agreements comprised between 77 and 89%. Discrepancies between IgE antibody methods have already been observed in other comparison studies [14–19]. Of particular note, most of the discrepancies between the two methods were observed for samples of very low allergen-specific IgE concentration (<3.5 kU_A/L). One hypothesis is that the limited IgE capture capacity of the nanofluidic sensors used on the NFA comprising the two-dimensional solid phase on which the allergens are bound resulted in a lower IgE-binding capacity compared to the sponge-like structure of the RM. Furthermore, the short interaction time during the few minutes of the assay may be particularly unfavorable for samples of very low concentration. In addition, a potential competition between allergen-specific IgE and IgG may influence test results in samples where the IgE concentration is the lowest and/or the allergen-specific IgG concentration is the highest. It is also possible, as hypothesized elsewhere [20], that in the context of NFAs, high-avidity antibodies are preferentially captured compared to antibodies of lower avidities. In addition, slight structural differences between allergens (caused by the primary sequence, folding in heterologous expression system, modification caused by the coupling chemistry, and/or drying of the sensors) may also affect allergen-specific IgE binding in some samples. As each diagnostic method has unique characteristics, the results are not necessarily interchangeable.

SPT provides a rapid in vivo response of the patient to the intradermic exposure to minute quantities of the allergens. These assays are mostly performed with crude allergenic extracts containing a mixture of antigens and other proteins, present in different, possibly low, con-

centrations. The results of SPT may depend on the performer and product potencies, which are not yet standardized. Moreover, patients under certain medication, i.e., antihistaminic, or patients with particular skin disorders (dermographism, atopic dermatitis, for instance) are not eligible for this method. Direct comparison with allergen-specific IgE assays, which measure soluble, free IgE, while SPT provides a measure of the skin response of clustered mast cell-bound, allergen-specific IgE, must therefore be interpreted cautiously. Results found in this study cannot easily be compared with other studies that performed similar comparisons. The present study enrolled only patients for whom clinical diagnosis could not be made according to patient's history and SPT. As a consequence, this patient population represented a difficult-to-diagnose group, possibly explaining a relatively lower percent positive and negative agreement than observed in pure analytical studies.

Indeed, in our study, the overall agreements of SPT versus IgE antibody assays were comprised between 67 and 90% for the RM and between 55 and 72% for the NFA, which is lower than the 85–95% range indicated elsewhere [21].

In addition, the use of single major allergens from house dust mite *D. pteronyssinus* or Timothy grass (*Phleum pratense*) pollen may not be representative of the profile of all allergic patients to house dust mites or grasses, since they may have been sensitized to other major or even minor allergens not included in our tests. It is therefore not surprising that testing only one component of these species is not sufficient to reach a high agreement with extract-based solution used in SPTs. On the other hand, higher agreements are observed for both the NFA and the RM with Bet v 1 and Fel d 1, which are the single major components of Birch tree (*Betula verrucosa*) pollen and cat (*Felis domesticus*) dander.

It is remarkable that the final clinical diagnosis is similar for the large majority of patients, as agreements were comprised between 92.4 and 99.1%, and despite a lower overall percentage agreement. This points out the partially limited impact of serological assays on the final clinical decision, which confirms that IgE serological assays are complementary to patient history and SPT.

Several methods are available to determine sIgE level, each of them using different technologies and allergen sources. As a consequence, test results vary from system to system [9]. In addition, allergen-specific IgE levels are today almost exclusively measured on large analyzers in clinical laboratories, which results in a total turnover time of several days. Moreover, in countries where ac-

cess to IgE assays in clinical laboratories is difficult, easy-to-use, precise, and accurate near-patient in vitro diagnostic platforms may spread the measurement of serum IgE. Whereas point-of-care tests are today widely available and routinely used for many analytes in various medical fields, as exemplified by blood gas analyses in hospital intensive care units, D-dimer measurements in emergency departments, or glucose testing in primary care offices, allergists do not yet have access to affordable, easy-to-use, precise, and accurate near-patient quantitative IgE assays. Quantifying allergen-specific IgE levels at the point of care would therefore allow allergists to obtain patient history, SPT results, and serological assay results within a single consultation.

In conclusion, in a difficult-to-diagnose population, a good analytical agreement was found between the two methods. More importantly, the agreement in clinical diagnoses was found to be excellent, which clearly highlights that allergen-specific IgE assays on NFAs are clinically as effective as the use of similar tests performed in a laboratory. Further clinical and economic impact studies will add more information on the global benefit of performing IgE serological assays at the point of care. Furthermore, additional allergy tests covering allergists' needs worldwide will broaden the utility of this novel nanofluidic system.

Statement of Ethics

The study was approved by Commission cantonale (VD) d'éthique de la recherche sur l'être humain, protocol 309/12, and registered in www.clinicaltrials.gov under reference NCT03599908. Written informed consent was obtained from all subjects before participating in the study.

Disclosure Statement

Fabien Rebeaud and Laurie D. Girard have stock options in Abionic SA. Iwan Maerki is a common shareholder. All other authors have no conflicts of interest to declare.

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Author Contributions

François Spertini, Samuel Roethlisberger, Fabien Rebeaud, Iwan Maerki: conceived clinical study design. Daria Mapelli, Ré-gine Audran, Vincent Aubert: data collection. Laurie D. Girard, Fabien Rebeaud, Iwan Maerki, Samuel Roethlisberger: data analysis. François Spertini, Samuel Roethlisberger, Fabien Rebeaud, Iwan Maerki, Laurie D. Girard: interpretation of the results. Annette Leimgruber, Guillaume Buss, Joël Duc, Felicitas Langner-Viviani: allergy committee expert members. All authors read and approved the manuscript.

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