

REVIEW ARTICLE

Prick test: evolution towards automated reading

X. Justo¹, I. Díaz¹, J. J. Gil¹ & G. Gastaminza²

¹CEIT and TECNUN, University of Navarra, San Sebastián, Spain; ²Department of Allergology and Clinical Immunology, Clínica Universidad de Navarra, University of Navarra, Pamplona, Spain

To cite this article: Justo X, Díaz I, Gil JJ, Gastaminza G. Prick test: evolution towards automated reading. *Allergy* 2016; **71**: 1095–1102

Keywords

allergic reaction; prick test; wheal; automatic measurement.

Correspondence

Gabriel Gastaminza, Department of Allergology and Clinical Immunology, CUN, University of Navarra, Av. Pío XII, 36, E-31008, Pamplona, Spain.
Tel.: +34 948255400
Fax: +34 948296500
E-mail: gastaminza@unav.es

Accepted for publication 19 April 2016

DOI:10.1111/all.12921

Edited by: Werner Aberer

Abstract

The prick test is one of the most common medical methods for diagnosing allergies, and it has been carried out in a similar and laborious manner over many decades. In an attempt to standardize the reading of the test, many researchers have tried to automate the process of measuring the allergic reactions found by developing systems and algorithms based on multiple technologies. This work reviews the techniques for automatic wheal measurement with the aim of pointing out their advantages and disadvantages and the progress in the field. Furthermore, it provides a classification scheme for the different technologies applied. The works discussed herein provide evidence that significant challenges still exist for the development of an automatic wheal measurement system that not only helps allergists in their medical practice but also allows for the standardization of the reading and data exchange. As such, the aim of the work was to serve as guideline for the development of a proper and feasible system.

In vivo tests in allergy date back to 1867 when Dr. Charles Blackley applied a porridge of pollen in water on his scratched skin, causing redness, swelling and itching. Later, Schick and Cooke introduced the intracutaneous test. However, it was not until 1950 that Sir Thomas Lewis proposed administering the allergen via the puncture technique (1).

From the onset of using skin tests in the field of allergy, scientists have tried to standardize the reactions obtained so that they are comparable regardless of where and how they have been generated. In general, the objective of standardization covers three parts: the puncture device, the allergens and the process (test and diagnosis).

Regarding the process, the most commonly used method for detecting allergies is known as the prick test. This method is relatively simple and fast. However, prick test, as it is performed nowadays, has some limitations in its diagnostics accuracy, especially for food allergy diagnosis (2–4). Besides, the analysis and measurement of the size of the wheals that correspond to a positive reaction are performed in a manual and laborious manner, or by means of a qualitative judgement made by the allergist. While some allergists just visually inspect the reaction, or compare it with the histamine wheal, others try to quantitatively measure the geometry of the wheal.

In general, two main parameters are used: the mean diameter of the wheal and the area, which shows less variability (5). These results have to be interpreted by the allergist, which can introduce additional variability. It has been shown that experts examining the same contours can arrive at different conclusions (6–8). An automated device would potentially reduce the uncertainties introduced by the reading process, minimizing the variability after the puncture is carried out. As Poulsen et al. (9) stated, the two main sources of uncertainties during the reading process are (i) the drawing of the wheal and (ii) the determination of the area. The variability produced by these factors can be significantly reduced by applying new technologies. Even other sources of error not usually taken into account, such as a test misplacement or an incorrect allergen assignment, could be eradicated by using a proper reading method.

This review aimed to describe and analyse the different technologies in the literature that have attempted to automate and standardize the process of measuring the outcomes of an allergy test, pointing out their advantages and disadvantages and discussing what is actually needed in order to develop a reliable wheal measuring system. The focus of the paper is on the reading and measuring part of the outcomes of the prick test, but not on the process or the puncture.

Methods and systems for automatic wheal measurement

To assemble a comprehensive overview of existing systems and methods for automatic wheal measurements, we searched for systems presented in scientific publications and patents. Various methods of organization were considered, including first dates of publication and relative complexity, and we selected categorization by technology. Inside each category, papers were organized according to the year of publication.

The different published methods for measuring wheals can be grouped into six categories, depending on the technology they apply: (i) 2D scanner, (ii) blood flow, (iii) skin impedance, (iv) thermography, (v) photography and (vi) 3D scanner. Figure 1 shows the timeline for the reviewed publications grouped into the six categories.

2D scanner

With the advent of personal computers, scanning devices became more common. In 1982, Ownby (10) was the first author to use a computer in order to computerize the process of determining the area of the wheal. In the study, he presented a method for scanning the test sheet and calculating the area of the wheal with the computer. Results were compared with those obtained by measuring the weight of the paper portion covered by the wheal. The relationship between the reference area and the computed area was very good. The method was able to significantly reduce the time required to determine the area of the wheal, but it was not so efficient with regard to other parameters, such as the maximum and minimum diameters. The variability obtained by the computer was less than 6% for areas over 20 mm², and it was over 16% for smaller areas.

Later, Poulsen et al. (9) improved previous results thanks to more powerful computers and scanners which allowed scanning directly over the area of interest. In addition, Poulsen et al. studied both the variability that could be introduced by the program that computed the area and the variability that could arise just prior to the scanning process (Fig. 2): uncertainties introduced by the allergist (with

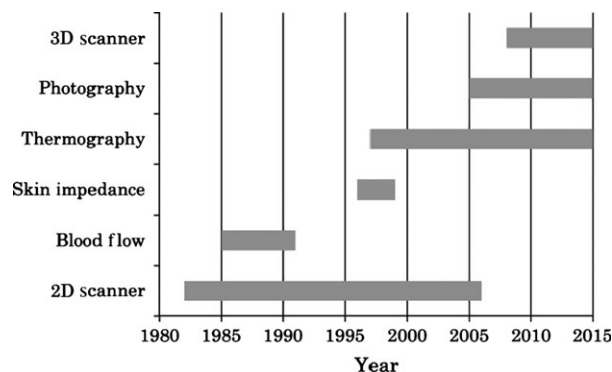


Figure 1 Emergence and development period of the different technologies for measuring wheals.

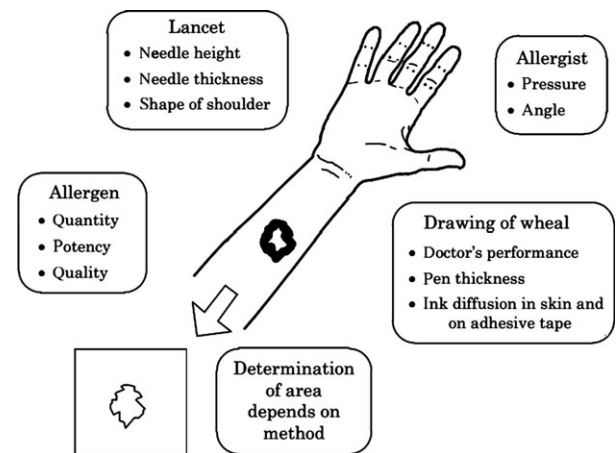


Figure 2 Factors affecting prick test results grouped into categories (picture adapted from (9)).

different pressure and/or angle when pricking the skin), the variability produced by different lancet models, the concentration of the allergen used as well as its quantity and quality, the precision in marking the wheal, the thickness of the marker and the spread of ink on the skin and on the tape. They also introduced a precalibration process for the scanner. Overall, the results improved upon previous systems. The variability in-between the same operator and among different operators remained around 2% and 3% on average, respectively. However, the most noticeable result was the measure of disturbance introduced by the doctor when marking the border of the wheal, which was beyond 4% for individual operators and nearly 6% for different operators, which is greater than the variability of the machine itself.

Three years later, Pijnenborg et al. (11) analysed the scanner devised by Poulsen in depth. The study looked more closely at the accuracy of the machine itself. It also analysed the variability between different operators and the same operator. The results were considered good enough (1.43% variability for intraoperator use, 1.85% day-to-day for the same operator and 2.34% for interoperator), although various limitations of the measurement process were noted: defining the exact contour is impossible due to the spread of ink first in the patient, then in the tape and finally in the paper, creating a blurred line in which it is difficult to determine where the border is actually located.

It was not until several years later that the process became more automated. Prinz et al. (12) proposed, based on previous studies, parallelizing the scanning of wheals to speed up the process. The method was based on the recognition of several markers that corrected position errors when scanning the test sheet so that when using a default template the program always knew what allergen corresponded to each contour. The accuracy of the calculated area was reduced for the sake of robustness and speed, because the system did not require a fully enclosed contour to determine the area boundary. This was achieved by applying a series of filters that blurred the outline until it became enclosed, obtaining the centreline

and fusing it with the original image to obtain an approximation of the wheal (Fig. 3). As a result, the variability rose up to 7%, but the system was able to calculate the area of up to 20 wheals in 20 min. The following year, the same team reviewed the edge detection algorithms, improving the accuracy of measuring the wheal area up to 99%, yielding a variability that was under 2% and reducing the processing time to 15 min (13).

This methodology almost reached its limits in 2006. It was still possible to reduce both the variability and the processing time (the latter thanks to advances in computers), but it continued to fail to address the main cause of errors, the variability of the prick test reading. As Poulsen stated in (9), the process needed for scanning the wheal contour introduces a lot more variability than the scan itself.

Blood flow

Laser Doppler flowmetry (LDF) can be used in hemodynamics research as a technique to partially quantify the blood flow in human tissues such as skin. The beam from a low-power laser penetrates the skin sufficiently to be scattered with a Doppler shift by the red blood cells and returns to be concentrated on a detector (Fig. 4).

Using this concept and previous work (14), Serup et al. (15) decided to determine the relationship between the wheal resulting from an allergic reaction and the blood flow around it. They tested several concentrations of histamine and sodium chloride in adults and measured the blood flow at different elapsed times and distances from the centre of the wheal. The results showed a clear relationship between the concentration of histamine and the flow measured at the periphery of the wheal. Interestingly, in the centre of the wheal, that is the puncture point, the blood flow measured was the same in all cases. This was because the flow caused by the puncture itself masked the flow caused by the wheal. The authors concluded that blood flow measurement had

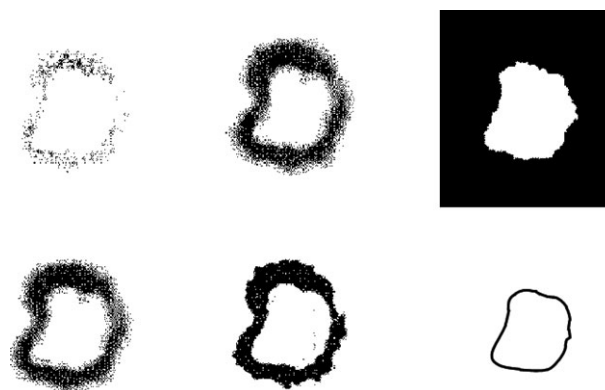


Figure 3 Contour restructuring process (from left to right and top to bottom): original contour, blurring, closed perimeter detection, overlay the contour with the original, contrast increase and final detection (picture adapted from (12)).

many benefits compared to the conventional prick test. There was still great variability in terms of allergen concentration, puncture and lancet type used, but the variability of the reading process could be significantly reduced. In contrast, the variability of the results was not analysed.

In a later work, Hovell et al. (16) analysed the system used by Serup in detail. They showed that the measurement performed exactly at the point of puncture was inconclusive, showing the maximum difference at one centimetre away and gradually decreasing to 5 cm, where it merged again with the background flow. Thus, it was concluded that the measurement should be performed as close to the point of incision as possible, but not over it. This study demonstrated that the blood flow measure for wheal characterization is possible and can eliminate the uncertainty introduced by the doctor when directly scanning or measuring the wheal, but there are also new uncertainties in this method: the distance at which

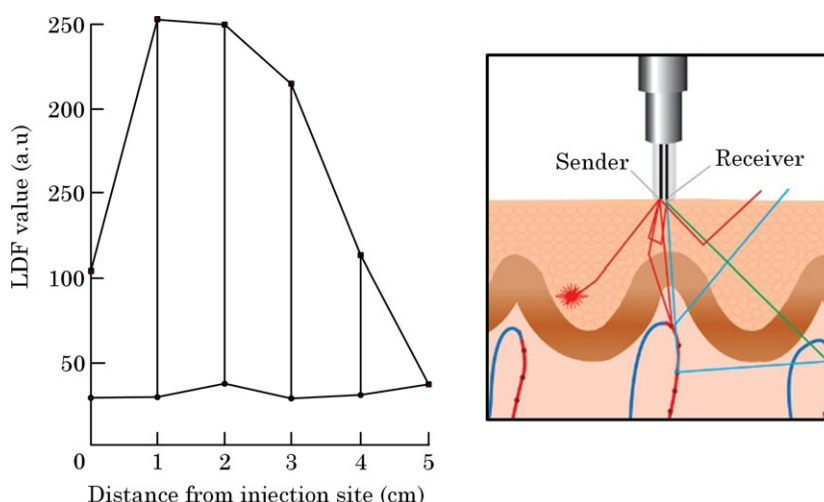


Figure 4 Blood flow measured as a function of distance from the injection site (left) (16). Schematic operation of a laser Doppler flowmeter (right).

the flow is measured, the perpendicularity of the measuring device (which affects the results) as well as the time elapsed since the puncture.

Subsequent studies (17–19) have conducted more experiments on this technique, obtaining slightly better results. In summary, this technology has some advantages in terms of reading accuracy, but it also adds new sources of variability through the process. Additionally, the high cost of the flowmeters caused this technique to fall into disuse in allergy.

Skin impedance

Biological tissue has electrical characteristics that can be gathered as an impedance. This parameter is difficult to represent; therefore, a set of four indices are extracted: the magnitude index, the phase index, the real part index and imaginary part index.

Nyren et al. (20) performed several tests based on this idea. The method had already been applied to the study of eczema and skin reactions, but its application to allergic wheals had not yet been carried out. Tests were conducted over 10 allergic volunteers by administering 40 µl of the allergen to which they were sensitive and histamine on the middle of the forearm at opposite sides. The relative position between the allergen and histamine was randomized. Results were measured in three ways: visual inspection (with a score of 1–4 relative to histamine), blood flow and skin impedance. The results showed a relationship between the visual and blood flow measures. A relationship was also found in the phase index of the impedance spectra, but not with the other indices. The variability of the results was not analysed.

Later, the same authors (21) conducted a study on the application of the skin impedance technology in the field of dermatology, concluding that the method could be very interesting for dermatitis. However, they also stated that its application to allergic wheal measurement, although possible, was not as promising as blood flow. Moreover, there were no advantages in terms of equipment cost or usability.

Thermography

This technology is based on the detection of radiation in the infrared range of the electromagnetic spectrum to produce images of that radiation. Because the amount of radiation emitted by an object increases with the temperature, thermography allows to graphically represent variations in temperature. Its application to allergy comes from the fact that allergic reactions cause the dilation of capillaries, increasing blood flow, which results in a temperature variation (Fig. 5).

The first articles on this technology studied the relationship between the area of erythema and the area obtained by thermography (22–24), but it was not until the late twentieth century when Bagnato et al. (25) used thermography to analyse allergic wheals. They conducted a study with 26 subjects: 16 of them sensitive to Parietaria and 10 healthy nonatopic subjects. The procedure was standard: a drop of allergic extract of Parietaria or a negative control was placed randomly on the forearm skin of the subjects with a standardized needle and moderate pressure. A baseline thermography image of the forearm was taken prior to testing and another image was taken 15 min after pricking. The difference in mean temperature between the post-testing and the baseline detection was measured. This allowed a correlation between the mean wheal diameter and changes in skin temperature to be found.

The results showed a correlation close to 94% and a variability below 5% in several tests carried out on different days with the same volunteers. The authors claimed that the system was useful for studies that require a greater degree of precision in evaluating skin reactivity, but for common diagnosis purposes, it turned out to be expensive and complex.

Later, Rokita et al. (26) obtained an equation that related skin temperature variations to the size of the reaction and the time elapsed after pricking (27). However, these variations were related to allergic sensitivity within a 0–5 scale relative to histamine. These measures are not as powerful as other parameters such as the medium diameters or the area.

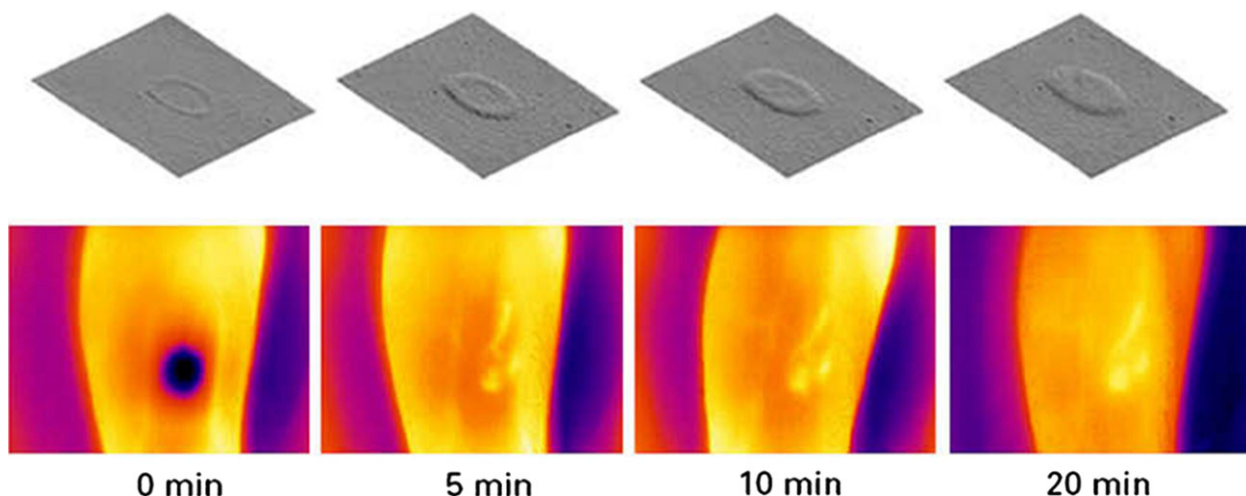


Figure 5 Evolution of a wheal over time after pricking in 3D (above) and by a thermographic camera (below) (picture adapted from (34)).

Currently, the diagnosis of allergies by means of thermography is in development and shows promising results. However, the evaluation parameters obtained by this technology differ from the ones currently used in common practice. This does not mean that the parameters obtained by this technology are neither wrong nor less accurate.

Photography

This system consists of a digital camera, a structure to support it and postprocessing software that allows the picture to be studied in order to obtain the contour of the wheal (Fig. 6). The first works only calculated the area of erythema caused by the allergic reaction (28), but nowadays it is also possible to calculate the area of the wheal.

At the beginning of the 21st century, Suchecki et al. (29) used a digital camera at a distance of 10 cm to take a picture of the forearm and the back. Lighting is a very important factor for image processing, because the colours that the sensor captures depend on it. Therefore, Suchecki et al. tried two variants of lighting: halogen and incandescent. For data processing, they used the HSV (hue saturation value) representation model. The area of the erythema and the wheal

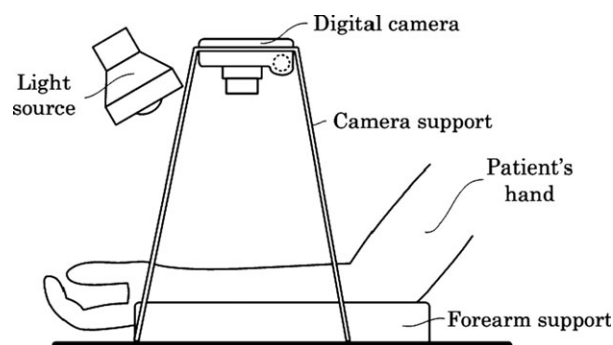


Figure 6 Assembly diagram of a photographic wheal identification system comprising a light source, a camera and a support structure.

was estimated by analysing histograms of HSV components. Contours of objects were estimated using median filtering due to the high noise usually found in such images.

Later, dos Santos et al. (30) performed a comprehensive study with 80 volunteers (31 men and 49 women) aged between 4 and 67 years. For all tests, a drop of histamine, *Dermatophagoides pteronyssinus* or saline solution was applied at the forearm, and digital photographs were taken at 0, 3, 5, 10, 15 and 20 min after the test was initiated. The wheal area was selected and ascertained using two Adobe Photoshop® filters: a plastic wrap and poster edges (Fig. 7). Then, pixels within the contour were correlated with mm². The method was compared with common ruler and adhesive tape reading methods to measure the mean diameter and contour of the wheal. Results showed a correlation of 0.54 between the digital approach and the mean diameter measure and a correlation of 0.71 with the contour. The intraobserver and interobserver variabilities of the digital method were 0.5% and 6%, respectively.

In the last decade, Bulan (31) has developed this concept in line with new technologies. The basic principle is the same, but the system is intended for a wider audience by using the camera of a smartphone. The method operates by first localizing the test region from captured images by detecting the calibration marks previously drawn on the skin. Then, a colour transformation from RGB to YCbCr (Y refers to luminance and Cb-Cr to chrominance) is performed and the luminance channel Y is discarded to eliminate illumination variation across the test region. With further processing, the method obtains an image with enhanced contrast where it is possible to detect the wheal. It is a slightly different method from previous works, but it includes a key point, which is the removal of the luminance, and thus, there is no dependence on the quantity and quality of light. In contrast, the accuracy for calculating the area of the wheal is reduced due to the lower quality of smartphone cameras.

As a general conclusion, reading systems based on photography offer an inexpensive option to increase the precision of the prick test. However, they do not meet yet the standards

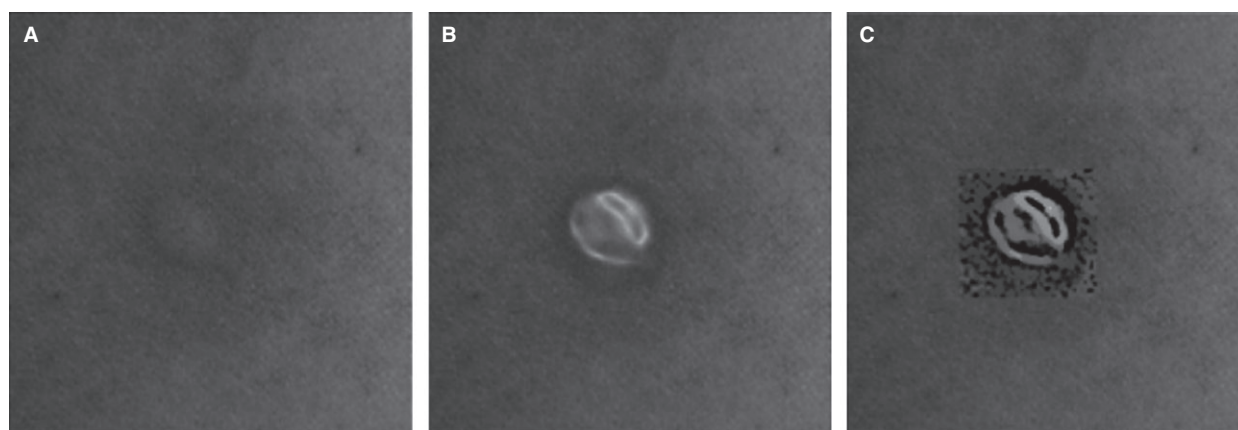


Figure 7 Recognition process of the wheal: (A) original photograph, (B) after plastic wrap filter and (C) after poster edges filter (picture adapted from (30)).

that allergists require in terms of accuracy. Moreover, we have not found any work that compares the measures obtained by photography with other methods and technology reviewed in this paper, such as thermography or blood flow. Another drawback of this technology is that it is very difficult to detect contours and measure wheals in black people because colour contrast is much lower in these cases.

3D scanner

It is a brand new technology in the field of allergy. In addition to computing the area and the maximum, minimum and mean diameters, 3D image sensors also provide data to determine both the height and the volume of a wheal.

The surface of an object can be digitalized by using several technologies and devices, but the most important ones can be grouped into two: 3D scanners based on triangulation and based on projecting structured light. The first technique shines a light beam (usually a laser) on the object and uses a camera sensor to capture its reflection. The camera is located at a known distance and angle with respect to the light beam. Thus, triangulation can be applied to precisely locate the object. The latter technique projects a pattern of light on the surface of the object and a camera looks at its deformation and shape to calculate the relative position of every point in the field of view.

With regard to this technology, dos Santos et al. (32) studied both the accuracy and consistency of using a commercial 3D acquisition system for wheal detection. The apparatus was previously calibrated with a wheal model of 50 mm². In their work, they obtained very little intraoperator and inter-operator variability for repeated measurements of the volume (0.74%) and area (0.37%). The results also showed superior accuracy over traditional ruler measurements for assessing prick test reactions to histamine and allergen (Fig. 8).

This technology presents an advantage over the previous ones, because it is possible to obtain the volume of the papule. Yet this parameter may not be useful because it has not been proved that it correlates with any diagnosis measure. Nevertheless, this technology allows to measure the diameters and the area of the wheal with high accuracy. Moreover, skin prick testing in black people is possible,

because this method is not based on finding colour contrast for detecting the wheal contour as in digital photography. However, the work by dos Santos et al. analyses each wheal individually, while current needs in medical practice require an automated system able to simultaneously capture and process several reactions to allergens.

Discussion and conclusions

An initial discussion point could be whether the development of a system to improve the measurement accuracy of the prick test makes sense having molecular diagnosis as an option. However, the prick test is nowadays the best, cheapest and most immediate system for performing a fast diagnosis of allergies. Moreover, prick tests performed using purified allergens as profilin or lipid-transfer protein have demonstrated similar sensitivity and specificity compared to *in vitro* determination (33).

On this basis, this paper has reviewed different attempts found in the literature to automate the process of reading the results of a skin prick test, concluding that up to date no standardized automatic system exists for recording and interpreting skin prick tests, although several methods and devices have been developed in the last decades. The outcomes of the prick test usually lack precision and are subjected to multiple sources of variability. Moreover, the measurement is very laborious and time-consuming.

If we were asked for the ideal specifications of a system able to automatically measure the outcomes of the prick test, in our opinion they could be mainly summarized by a system able to provide a quantitative and precise diagnosis in the same time required for a visual qualitative inspection. Moving into more detail, the system should be:

- Fast: letting as little time as possible to lapse between pressing a button and obtaining the measurements on the screen.
- Precise: the geometry of the wheal, maximum and mean diameters, contour, area and even height should be measured with the precision of micrometres.
- Simple: it should not be necessary to undergo a long practice period to manage the system.

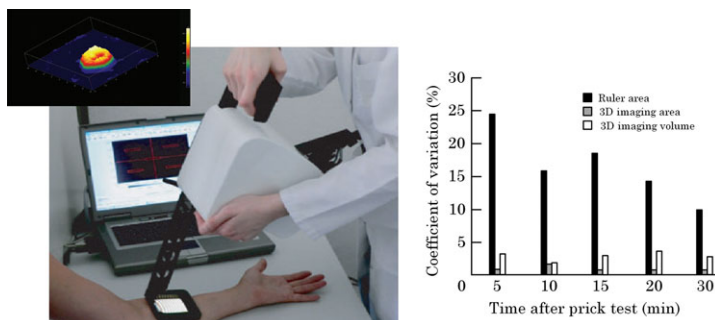


Figure 8 3D acquisition system applied to wheal detection (left). Comparison of the variability obtained with different measuring methods (right) (picture adapted from (32)).

- Traceable: databases and medical history should be easily created to precisely compare the within-patient outcomes over time, or even between patients.
- Affordable: it is difficult to put a market price on such a system, but it should not be very expensive because the current art does not require any machine.

Most of the systems described in this paper are not practical for use in a busy clinical practice due to the high cost of the equipment and the time needed to process the results. Besides, most of them still lack sufficient precision and low variability levels. Therefore, it is necessary to develop a system that is able to perform a simple and rapid diagnosis with reliable and accurate results.

Having analysed the different technologies, it seems that future developments in photography and 3D scanners may pave the way to a reliable wheal measurement method.

Furthermore, measuring the volume and height of the wheal may open new means of diagnosis. However, this technology still has to deal with several challenges: simultaneous measurement of multiple wheals, fast computer processing of data and the development of a simple and low-cost device.

Author contributions

All the authors have collaborated in conception and design of the study, in drafting the article, bibliographic search and have approved its final version.

Conflict of interest

The authors declare that they have no conflicts of interest. No funding has been received for this manuscript.

References

- Bernstein IL, Li JT, Bernstein DI, Hamilton R, Spector SL, Tan R et al. Allergy diagnostic testing: an updated practice parameter. *Ann Allergy Asthma Immunol* 2008;**100**:S1–S148.
- Soares-Weiser K, Takwoingi Y, Panesar SS, Muraro A, Werfel T, Hoffmann-Sommergruber K et al. The diagnosis of food allergy: a systematic review and meta-analysis. *Allergy* 2014;**69**:76–86.
- Michel S, Scherer K, Heijnen IAFM, Bircher AJ. Skin prick test and basophil reactivity to cetuximab in patients with IgE to alpha-gal and allergy to red meat. *Allergy* 2014;**69**:403–405.
- Vlieg-Boerstra BJ, van de Weg WE, van der Heide S, Dubois AEJ. Where to prick the apple for skin testing? *Allergy* 2013;**68**:1196–1198.
- Vohlonen I, Terho EO, Koivikko A, Vanto T, Holmen A, Heinonen OP. Reproducibility of the skin prick test. *Allergy* 1989;**44**:525–531.
- McCann WA, Ownby DR. The reproducibility of the allergy skin test scoring and interpretation by board-certified/board-eligible allergist. *Ann Allergy Asthma Immunol* 2002;**89**:368–371.
- Oppenheimer J, Nelson H. Skin testing: a survey of allergists. *Ann Allergy Asthma Immunol* 2006;**96**:19–23.
- Berardesca E, Gabba P, Nume A, Rabbiosi G, Maibach H. Objective prick test evaluation: noninvasive techniques. *Acta Derm Venereol* 1992;**72**:261–263.
- Poulsen LK, Liisberg C, Bindslev-Jensen C, Malling HJ. Precise area determination of skin-prick tests: validation of a scanning device and software for a personal computer. *Clin Exp Allergy* 1993;**23**:61–68.
- Ownby DR. Computerized measurement of allergen-induced skin reactions. *J Allergy Clin Immunol* 1982;**69**:536–538.
- Pijnenborg H, Milsson L, Dreborg S. Estimation of skin prick test reactions with a scanning program. *Allergy* 1996;**51**:782–788.
- Prinz M, Vigl K, Wohrl S. Automatic measurement of skin wheals provoked by skin prick tests. *Stud Health Technol Inform* 2005;**116**:441–446.
- Wohrl S, Vigl K, Binder M, Stingl G, Prinz M. Automated measurement of skin prick tests: an advance towards exact calculation of wheal size. *Exp Dermatol* 2006;**15**:119–124.
- Bisgaard H, Kristensen JK. Quantification of microcirculatory blood flow changes in human cutaneous tissue induced by inflammatory mediators. *J Invest Dermatol* 1984;**83**:184–187.
- Serup J, Staberg B. Quantification of wheal reactions with laser Doppler flowmetry: comparative blood flow measurements of the oedematous centre and the perilesional flare of skin-prick histamine wheals. *Allergy* 1985;**40**:233–237.
- Hovell CJ, Beasley RW, Mani R, Holgate ST. Laser Doppler flowmetry for determining changes in cutaneous blood flow following intradermal injection of histamine. *Clin Allergy* 1987;**17**:469–479.
- Olsson P, Hammarlund A, Pipkorn U. Wheal and flare reactions induced by allergen and histamine: evaluation of blood flow with laser Doppler flowmetry. *J Allergy Clin Immunol* 1988;**82**:291–296.
- Wa TCLK, Almond NE, Cooke ED, Turner P. Skin blood flow changes following intradermal Bradykinin injections measured by laser Doppler flowmetry: comparison with wheal and flare. *J Med Eng Technol* 1990;**14**:190–193.
- van Neste D. Skin response to histamine: reproducibility study of the dry skin prick test method and of the evaluation of microvascular changes with laser Doppler flowmetry. *Acta Derm Venereol* 1991;**71**:25–28.
- Nyren M, Ollmar S, Nicander I, Emtestam L. An electrical impedance technique for assessment of wheals. *Allergy* 1996;**51**:923–926.
- Nyren M, Hagstromer L, Emtestam L. On assessment of skin reactivity using electrical impedance. *Ann N Y Acad Sci* 1999;**873**:214–220.
- Phipatanakul CS, Slavin RG. Use of thermography in clinical allergy. *J Allergy Clin Immunol* 1972;**50**:264–275.
- Stuttgen G, Flesh U, Witt H, Wendt H. Thermographic analysis of skin test reactions using AGA thermovision. *Arch Dermatol Res* 1980;**268**:113–128.
- Uematsu T, Takiguchi Y, Mizuno A, Sogabe K, Nakashima M. Application of thermography to the evaluation of the histamine skin test in man. *J Pharmacol Methods* 1987;**18**:103–110.
- Bagnato GF, Gulli S, Denuzzo G, di Cesare E, Sansotta C. Measurement of allergen-induced skin reactions by computerized dynamic telethermography. *J Invest Allergol Clin Immunol* 1997;**7**:238–241.
- Rokita E, Rok T, Taton G. Thermographic measurements of allergen-induced skin reactions. *World Congress Med Phys Biomed Engineering* 2009;**13**:120–123.
- Rokita E, Rok T, Taton G. Application of thermography for the assessment of allergen-induced skin reactions. *Med Phys* 2011;**38**:765–772.
- Setaro M, Sparavigna A. Quantification of erythema using digital camera and computer-based colour image analysis: a multicentre study. *Skin Res Technol* 2002;**8**:84–88.

29. Suchecki R, Lipiec A, Wierzejski W, Zaworski W, Grzanka A. Computer planimetry in allergology skin prick tests. *Proceedings of the SPIE* 5775, Photonics Applications in Astronomy, Communications, Industry, and High-Energy Physics Experiments III 2005;**5775**:320–325.
30. dos Santos RV, Titus RG, Lima HC. Objective evaluation of skin prick test reactions using digital photography. *Skin Res Technol* 2007;**13**:148–153.
31. Bulan O. Improved wheal detection from skin prick test images. *Proceedings of the Image Processing Machine Vision Applications VII* 2014;90240J–90240J–10.
32. dos Santos RV, Mlynek A, Lima HC, Martus P, Maurer M. Beyond flat weals: validation of a three-dimensional imaging technology that will improve skin allergy research. *Clin Exp Dermatol* 2008;**33**:772–775.
33. Berroa F, Gastaminza G, Saiz N, Azofra J, Gamboa PM, Vela C et al. Invivo and invitro techniques in the diagnosis of lipid transfer protein sensitization. *Ann Allergy Asthma Immunol* 2013;**111**:571–573.
34. Siebenhaar F, Degener F, Zuberbier T, Martus P, Maurer M. High-dose desloratadine decreases wheal volume and improves cold provocation thresholds compared with standard-dose treatment in patients with acquired cold urticaria: a randomized, placebo-controlled, crossover study. *J Allergy Clin Immunol* 2009;**123**:672–679.