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REVIEW

Assessment of sudomotor function

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Abstract

Purpose To review the currently available literature on clinical autonomic tests of sudomotor function.

Methods We searched PubMED/MEDLINE for articles on technical principles and clinical applications of sudomotor tests with a focus on their drawbacks and perspectives in order to provide a narrative review.

Results The quantitative sudomotor axon reflex sweat test (QSART) is the most widely used test of sudomotor function. The technique captures pathology with low intra- and inter-subject variability but is limited by technical demands. The thermoregulatory sweat test comprises topographic sweat pattern analysis of the ventral skin surface and allows differentiating preganglionic from postganglionic sudomotor damage when combined with a small fiber test such as QSART. The sympathetic skin response also belongs to the more established techniques and is used in lie detection systems due to its high sensitivity for sudomotor responses to emotional stimuli. However, its clinical utility is limited by high variability of measurements, both within and between subjects. Newer and, therefore, less widely established techniques include silicone impressions, quantitative direct and indirect axon reflex testing, sensitive sweat test, and measurement of electrochemical skin conductance. The spoon test does not allow a quantitative assessment of the sweat response but can be used as bedside-screening tool of sudomotor dysfunction.

Conclusion While new autonomic sudomotor function testings have been developed and studied over the past decades, the most were well-studied and established techniques QSART and TST remain the gold standard of sudomotor assessment. Combining these techniques allows for sophisticated analysis of neurally mediated sudomotor impairment. However, newer techniques display potential to complement gold standard techniques to further improve their precision and diagnostic value.

Keywords Sweat · Neuropathy · Small fiber · Sympathetic · Autonomic

Introduction

Clinical evaluation of sudomotor function has become a standard component of clinical autonomic function testing which examines the integrity of the cholinergic part of the sympathetic nervous system. Over the past four decades, sudomotor tests have been shown to detect and quantify sudomotor dysfunction in various diseases such as small fiber neuropathies and neurodegenerative disorders [1–3]. Their application extends from diagnostic and localization of the disturbance to monitoring disease progression [4].

The two major thermoregulatory mechanisms of the human body are peripheral vasodilatation (vasoconstriction, respectively) of small skin vessels and sweat production to maintain a constant body temperature of 37 °C, in which the complex biochemistry systems of the human body work at their highest efficacy. The thermoregulatory center is the hypothalamus, which integrates afferent signals from peripheral and visceral thermoreceptors and maintains the core body temperature via two efferent pathways: somatic and sympathetic nerve fibers. Activation of these fibers results either in an increase, e.g., by muscle shivering, or decrease, e.g., by sweat production, of body temperature [5, 6]. The dermally located eccrine sweat glands are primarily

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innervated by unmyelinated postganglionic sympathetic C-fibers and are activated by acetylcholine [7]. If fully activated, the total of human sweat glands produces up to 3.5 L of sweat per hour [5]. Neural control of sweat production is largely influenced by environmental factors, such as temperature, humidity and is also age and gender dependent. This susceptibility leads to high environmental and technical demands of sudomotor function assessment [6].

It should be noted, that there are normal variations of sweat distribution. The sweat response is not only depending on the body area, but also may vary within body segments [8]. Different sweat gland densities, secretion rates, sensitivity to core and skin temperature changes and varying activation thresholds of sudomotor fibers, are among accepted explanations of topographical intra-subject variability [8]. The highest sweat gland density has been reported at the forehead, palmar and plantar with a caudal-to-rostral pattern of sweat onset [8].

Either an increased or a decreased sweating after adequate stimulation can indicate impaired sudomotor function and can become clinically relevant. Sudomotor dysfunction is a manifestation of autonomic peripheral neuropathies, a group of neurological disorders which selectively affects unmyelinated and small, lightly myelinated nerve fibers [2]. Diabetes mellitus constitutes the most frequent underlying disease of autonomic peripheral neuropathies in the western countries. Further causes of autonomic peripheral neuropathies include acute infections, both primary and hereditary amyloidosis, neoplasia (e.g., Lambert-Eaton myasthenic syndrome) and exposition to neurotoxins (e.g., Cisplatin) [2].

Patients with sudomotor function are clinically presenting either with increased sensation of sweating during higher environmental temperatures or with heat intolerance due to anhidrosis. Both types of sudomotor dysfunction can lead to significant limitations of life quality, expressed in e.g., avoiding high temperatures during summer by only staying in air-conditioned rooms or social interactions because of emotional or social embarrassments associated with extensive sweating. This highlights the importance of obtaining detailed medical history such as known medical disorders, list of medications and symptoms of autonomic disorders in patients with sudomotor disturbances. Moreover, thorough analysis of patient's habits and social life is important in the clinical work up. Patients may also experience minor changes of epidermal moisturization, such as hyperkeratosis and rhagades up to ulcers, which can be detected by the patient or more commonly by the examining physician. During the basic clinical evaluation of patients with suspected sudomotor dysfunction, the lower limbs are of special interest, as they are often uninfluenced by the patients daily dermal care cosmetic and therefore show inspectable dermal alterations. Therefore, inspection of the patients' socks and shoes (e.g., intense odor or excessive dander residues) can be helpful, to detect squealing symptoms of sudomotor dysfunction.

We reviewed the current literature on the most frequently used sudomotor function testing procedures, starting with the gold standards techniques [thermoregulatory sweat testing (TST) and quantitative sudomotor axon reflex sweat test (QSART)], followed by clinical tests needing further future clinical evaluation or having been shown to provide vague indications of sudomotor dysfunction (sensitive sweat test (SST), spoon test). Each clinical test of sudomotor function is presented with respect to its neurophysiological background, conduction, limitations and future perspectives.

Methods

We performed a narrative review. We searched the National Library of Medicine (MEDLINE) database as well as Google Scholar using the search terms "sudomotor", "sweat", "Thermoregulatory sweat testing", "Quantitative sudomotor axon reflex sweat test", "Silicone imprint", "Quantitative direct and indirect test of sudomotor function", "Electrochemical skin conductance", "Sensitive sweat test", "Sympathetic skin response", "Spoon test", "TST", "QSART", "QDIRT", "ESC", "SST", "SSR" as well as their combinations using the Boolean operators "AND" and "OR". Our literature search included studies from the first data available until the last search conducted in January 2018. Language restriction was applied including only articles in English.

Thermoregulatory sweat testing

The TST is the current gold standard to objectify the general, more specific pre- and postganglionic sudomotor function, of the ventral body surface. As a pioneer, Guttmann published a full description of the technique using Quinizarin as color indicator, summarizing 10 years of experience and development of the TST in 1947 [9]. The TST is performed in a humidity controlled (35–40%) and to 45–50 °C preheated room. The patient is nearly completely unclothed and lies in supine position on a testing table [1]. An indicator dye, which shows a pH change with a color change, is scattered on the complete ventral skin surface (omitting the eye, ears and perioral region). Skin and core temperature are measured repeatedly, whereas the aimed skin temperature is set to 38.5–39.5 °C, and the aimed increase of the core temperature is 1 °C from baseline measured temperature, or an increase to 38 °C, alternatively. The maximum heating time is set to 70 min to avoid patient's hyperthermia with corresponding clinical symptoms [1, 10]. Digital pictures of the sweating pattern in respect to the change of the indicator dye are taken. In the TST analysis the measured anhidrotic

skin area is divided by the total skin area and multiplied by 100 [1]. A physiological finding in TST would be a symmetric sweating pattern all over the ventral body surface.

Neurophysiological background

The peripheral thermoreceptors are signaling an increased mean skin temperature and blood temperature via the spinothalamocortical tract and lateral spinal cord to the thalamus and hypothalamus [11]. The preoptic-anterior hypothalamus area detects the core temperature and processes both the peripheral temperature and the core temperature [11]. Efferent pathways travel from the hypothalamus via the pons (tegmentum) and the lateral reticular medulla to the intermediolateral column. Thence, the preganglionic cholinergic neurons of the intermediolateral column synapse in the paravertebral sympathetic ganglia with postganglionic sympathetic cholinergic sudomotor axons, which end at the sweat glands [1]. This central autonomic pathway with consecutive peripheral sudomotor activation is activated upon increasing body and core temperature during the TST protocol. Therefore, TST can show pathological sweat patterns due to both central and peripheral lesions within the sudomotor system.

Clinical implications

The TST allows the evaluation of pre- and postganglionic axonal integrity. Additionally, this technique allows the examination of sudomotor function of body areas, e.g., the fingers and toes, which are normally not tested in the other local sweat tests [e.g., QSART and Quantitative direct and indirect test of sudomotor function (QDIRT)] [1]. If combined with tests of the postganglionic sudomotor function, TST is useful in the differentiation of preganglionic lesions, as they are characterized by an abnormal TST and normal QSART, silicone imprints and QDIRT. However, in later stages of centrally caused sudomotor function, peripheral denervation might be caused by the lack of central efferent activation thus compromising QSART results as well. A major advantage of the TST technique is its capability to characterize sweat patterns topographically. In fact, some TST-detected sweat patterns are leading the way for diagnosis of highly prevalent neurological disorders, such as neuropathies, ganglionopathies or generalized autonomic failure (Fig. 1) [12].

Limitations

The major limitation of TST is its high technical demand. To date, the technique is only conductible and interpretable in highly specialized clinical settings, due to the high demanding testing protocol. Fully equipped TST chambers are available in a handful of specialized centers [1]. The patient may experience this sweat test as stressful due to the required time commitment.

Perspective

To address the high patient commitment in the TST, indication should be made after careful consideration of the underlying cause of sudomotor dysfunction. If there is no anamnestic evidence for central, thus preganglionic, alterations, local postganglionic sudomotor function tests should be performed in first-line. Industrial production of semi-automated TST chambers might help establishing the technique more widely.

Quantitative sudomotor axon reflex sweat test

In 1983, Low and colleagues introduced the QSART as a quantitative testing method of postganglionic sudomotor function over a restricted area [1, 13, 14]. The local sweat production is measured as a change of relative humidity over time in a sweat capsule during and after skin preparation

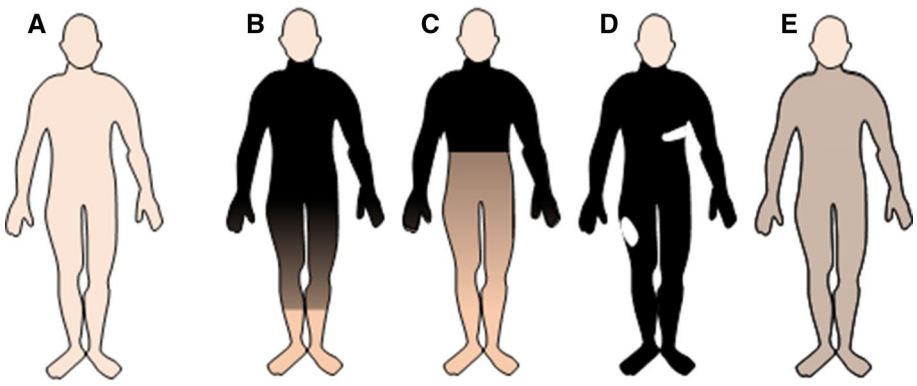

Image: Dr. Illigens and Dr. Siepmann, modified from [1].

Fig. 1 TST. The figure shows five exemplary findings on ventral body surface sweat pattern analyses on TST: normal sweat pattern (**a**). Diabetic neuropathy associated sweat pattern with stocking distribution (**b**). T8 myelopathy with consecutive sweat loss below the lesion (**c**). Left T5 radiculopathy and left lateral femoral cutaneous neuropathy (**d**). Sweat pattern in patients with pure autonomic failure (**e**) [3]

followed by iontophoresis of 10% acetylcholine [1, 14, 15]. The temporal resolution, latency, magnitude and duration of the sudomotor response are digitalized, plotted and analyzed using physiological response analysis software. Tested body sites are the forearm, proximal and distal leg and dorsum of the foot [1]. The sweat response starts gender-independent after a latency of 1–2 min in healthy controls, increasing until the maximum of sweat response is reached at approximately 5 min after stimulation [1]. Depending on the tested body site, the mean sweat output is in healthy females 0.25–1.2 µl/cm^2 and in males 2–3 µl/cm^2 [1].

Neurophysiological background

The QSART is based on the local spread of the sweat response via the axon-reflex (Fig. 2). Iontophoresis of acetylcholine is used to evoke the axon-reflex. Iontophoresis is a technique which utilizes an electric current to deliver charged molecules into the dermal skin layers. Iontophoresis of acetylcholine activates the terminal endings of the efferent sudomotor nerve fiber. More precisely, acetylcholine molecules bind to nicotinic and muscarinic receptors of the terminal nerve endings. Thus, a local sweat response is induced in the skin area of acetylcholine application (direct sweat response). Moreover, the binding of acetylcholine to nicotinic receptors at the sudomotor nerve terminals generates an action potential, which is antidromically transmitted until it reaches the first axon branch point [1]. There, the action potential is ephaptically transmitted to adjacent sudomotor nerve fibers and is then orthodromically conducted in these collateral efferent sudomotor nerve fibers, lastly evoking a sweat reaction in a so-called indirect skin area surrounding the direct skin area of acetylcholine application (indirect sweat response) [15].

Intensity of indirect sweating (relative humidity over time) indicates functional integrity of the nerve fiber mediating the axon-reflex sweat response.

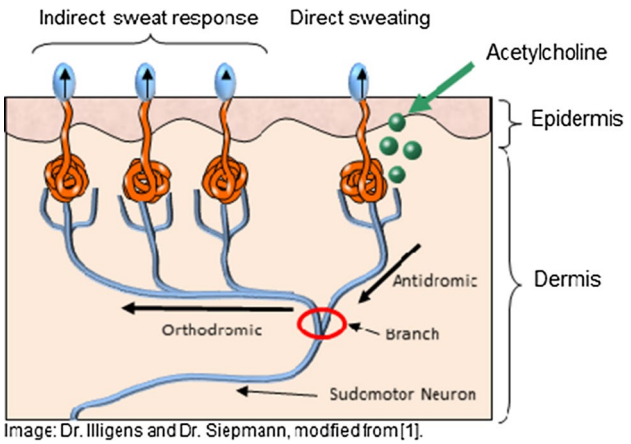

Fig. 2 Graphical illustration of the cholinergic sudomotor axon-reflex. Iontophoretic application of acetylcholine induces axon-reflex mediates sweating in a skin area which surrounds the area of iontophoresis. The reflex is mediated by unmyelinated sympathetic C-fibers. Upon stimulation of their terminal endings by acetylcholine, an action potential is generated which travels antidromically to an axon-branch point and then orthodromically to a neighboring population of sweat glands. Thus, an indirect sweat response is evoked in a skin area which surrounds the region to which acetylcholine has been applied

Clinical implications

QSART should be performed if a postganglionic sudomotor dysfunction is assumed. This method allows quantification of sudomotor function with respect of the temporal axon-reflex mediated sweat-response [16]. A reduced sweat volume in response to the stimulation with acetylcholine is the most frequent pathological observation. This finding may characterize a length-dependent neuropathy, particularly if the alteration lies predominantly distal [20]. However, an increased sudomotor sweat response may also be present in early stages of small fiber neuropathy, indicating supersensitivity of post-ganglionic nerve fibers after denervation. This may also be observed in reflex sympathetic dystrophy [4]. In combination with the TST, QSART may be helpful to discriminate between acute preganglionic and postganglionic denervation, as the former presents anhidrosis on the TST but shows unaltered response on QSART. Its discriminatory value is, however, restricted to the acute phase of the pathology, as long-established preganglionic dysfunction also results in reduced sweat response assessed via QSART. The strengths of QSART are its relatively low variability and high utility in detecting clinically relevant postganglionic small fiber dysfunction [1].

Limitations

Due to highly technical demands, QSART is currently not suitable for bedside sudomotor function testing. Several specific requirements to the testing laboratory and staff explain why QSART is mostly used in specialized autonomic centers. Importantly, the testing site needs to be controlled for temperature and humidity due to the impact of these factors on variability of measurement results. Moreover, the staff conducting assessment needs to be trained to ,e.g., guarantee stable measurement over time and avoid leaking of acetylcholine from the capsule [1]. Additionally, the QSART equipment is expensive and needs regular calibrations. Some patients may experience temporary discomfort due the iontophoresis procedure, such as burning sensation or skin irritation.

Perspective

Recent studies focused on the development of gel-based acetylcholine QSART-vehicles, aiming to reduce leaking from the iontophoresis capsule and patient discomfort during acetylcholine iontophoresis [17]. Conversely, the use of more flexible gum-based iontophoresis capsules might additionally reduce the leaking of acetylcholine. Moreover, it might be helpful if iontophoresis capsules were produced in a more suitable shape for each tested skin region. Strategies to facilitate the establishment of QSART outside specialized autonomic centers might be useful to increase the availability of postganglionic cholinergic assessment and thus improve patient care, e.g., in rural areas with limited access to university centers.

Silicone imprint

After iontophoresis of a cholinergic agonist (e.g., acetylcholine) and removal of the iontophoresis capsule, the skin is carefully dried. Afterwards, a thin layer of silicone is applied to the tested skin area and removed after complete polymerization, which takes depending on the used silicone type, approximately 5 min. To reach a higher plotting quality of the sweat droplet silicone imprints, toner can be applied to the silicone, followed by removing the toner from the unimprinted silicone surface with, e.g., alcohol swipes. The imprints can be analyzed either manually or using a software based semi-automated algorithm. Relevant parameters deriving from the silicone Imprint test include sweat droplet number, size and distribution. Thus, techniques allow detailed assessment of the axon-reflex mediated sweat response with spatial temporal resolution. However, it cannot capture the response over time. This differentiates it from QSART, a technique which evaluates the axon-reflex sweat response with temporal resolution. Standard droplet values are in healthy individuals 311 ± 38 sweat droplets/cm^2 in the hand (lower limit is set to 255 sweat droplets) and 281 ± 38 sweat droplets/cm^2 in the foot (lower limit is set to 235 sweat droplets) [1].

Neurophysiological background

Similarly, to QSART the silicone imprint method is based on an axon-reflex mediated local spread of the sweat response after iontophoresis of a cholinergic agonist as described in detail above [paragraph "QSART-Neurophysiological Background"]. However, the direct sweat response is also displayed with the silicone imprint method.

Clinical implications

The silicone imprint method can be used in the screening of patients with sudomotor dysfunction, as it is more economical and a less demanding technique compared with other available sudomotor function tests such as QSART. A reduced number of droplet impressions indicate the postganglionic sudomotor denervation, present in several autonomic neuropathies such as familial dysautonomia, diabetic neuropathy, Fabry's disease and congenital insensitivity to pain [18]. However, pathological sweat droplet imprints in this method are not specific for postganglionic sudomotor dysfunction, as a congenital lack of eccrine sweat glands or an occlusion of the excretory ducts of the sweat glands can also lead to the diagnosis of impaired sudomotor function.

Limitations

To avoid possible skin dander or hair imprints, the skin needs to be carefully prepared to enable an accurate analysis of the sweat droplets. Also, the utilized silicone can cause a biased impression of sudomotor function due to silicone reactions with the sweat droplets [14]. It is noteworthy that the silicone impression method is a rather time-consuming test because of the necessary post-hoc processing of each imprint. Additionally, the silicone imprint method does not allow a differentiation of direct and indirect sudomotor function.

Perspective

The establishment of silicone impressions as a standard screening test in clinical sudomotor function evaluation would require further standardization of the silicone material as well as the way it is applied to the skin. The silicone material should feature non-occlusive, hydrophobic and fast polymerization characteristics. A silicone which starts polymerization after contact with sweat components, like electrolytes or urea, might help increase precision of measurement results [1].

Quantitative direct and indirect test of sudomotor function

The QDIRT, developed by Gibbons and colleagues in 2008 evaluates functional integrity of postganglionic sudomotor function. Aiming to make sudomotor function testing outside of specialized clinical settings more feasible, the developers introduced a new analysis of sweat droplets by using an imaging analyzing software which is technically easier to perform than hygrometric assessment [14]. Similar to QSART, the QDIRT procedure comprises iontophoresis

of a 10% acetylcholine solution into the dermal layer of the skin to induce axon-reflex mediated sweating in an indirect skin area which surrounds the direct area of acetylcholine application. Before iontophoresis is initiated, the skin area is manually dried and completely covered with an indicator dye (e.g., povidone-iodine mixed with cornstarch and mineral oil for liquefaction). The color change of the used indicator marks the appearance of sweat droplets on the skin (Fig. 3). Color change due to axon-reflex sweating following iontophoresis of acetylcholine is assessed by repeated digital photography with pictures taken every 15 s over a general time of 7 min (Fig. 4). Thus, the QDIRT combines temporal and spatial analysis of axon-reflex mediated sweating which might improve precision of measurement when compared to techniques limited to spatial (e.g., silicone imprint) or temporal (e.g., QSART) resolution.

The direct stimulation testing area is defined by the skin surface with direct acetylcholine contact, whereas the area of the axon-reflex response (indirect area), is defined by the total diameter of iontophoresis capsule minus the diameter of the indirect iontophoresis area [14].

Sweat droplets are analyzed for their number, size and the percentage of sweat droplet change over a total area. Temporal analysis of sweating is obtained by determination of change in sweat area over the change in time [14].

Neurophysiological background

Similarly, to QSART and the silicone imprint method, QDIRT is based on an axon-reflex mediated local spread of the sweat response after iontophoresis of a cholinergic agonist as described in detail above [paragraph "QSART-Neurophysiological Background"].

Clinical implications

The QDIRT can be conducted in non-specialized clinical settings as the sudomotor function analysis comprises automated and semi-automated procedures and has comparatively low technical demands [1, 13]. Therefore, the QDIRT might be useful for sudomotor function testing in general neurology departments without an autonomic testing lab. However, the environmental requirements such as controlling for humidity are still considerable and, even more importantly, the QDIRT has been rarely used in research which explains the absence of any normative values clinical measurements could be compared with.

Limitations

As the quantification of sweat response in QDIRT is recorded by the change of the indicator dye, sweat production in areas were the indicator dye has already changed can be missed. In contrast to QSART the QDIRT does not display the sudomotor function over a predefined area, which may reduce the interindividual comparability of the QDIRT. Although the QDIRT measurement procedure comprises several semi-automated sudomotor function tests, it requires trained clinical staff as well as minimization of environmental influence factors such as changes in temperature or humidity. Moreover, the digital camera used to capture the sweat response needs to be adjusted properly to avoid light reflections, which requires thorough preparation. Therefore, the QDIRT would need to undergo technical advancements to be used as a bed side test. Similar to other sudomotor function tests utilizing iontophoresis, patients may experience temporary skin irritation. To date, few studies have

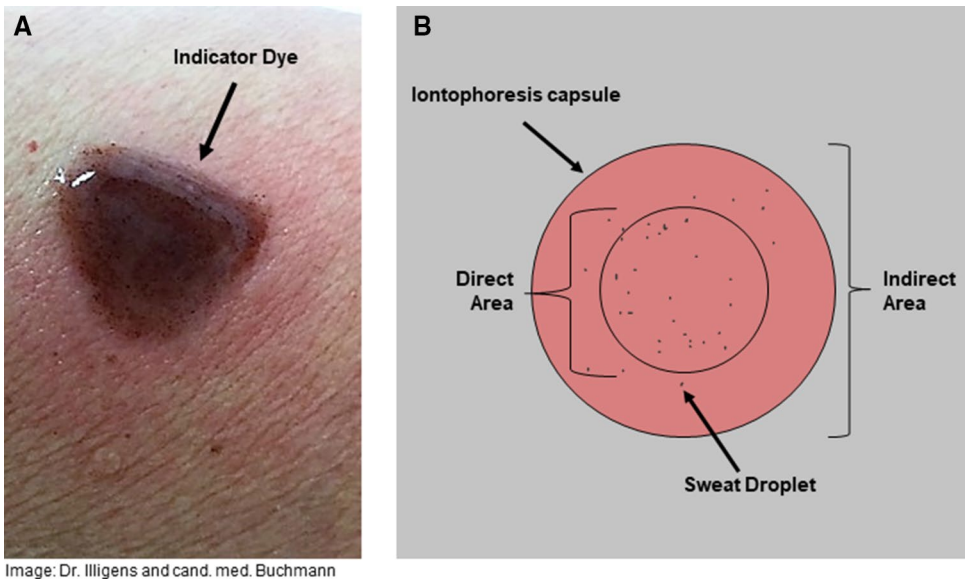

Fig. 3 QDIRT. The image shows a photograph of a skin region where axon-reflex sweating has been induced by iontophoresis of acetylcholine (**a**). The skin had been pretreated with indicator dye consisting of povidone-iodine, cornstarch and mineral oil to highlight sweat droplets. Axon-reflex mediated sweating is evaluated by quantifying sweat droplets in the indirect skin area surrounding the direct area of acetylcholine application (**b**)

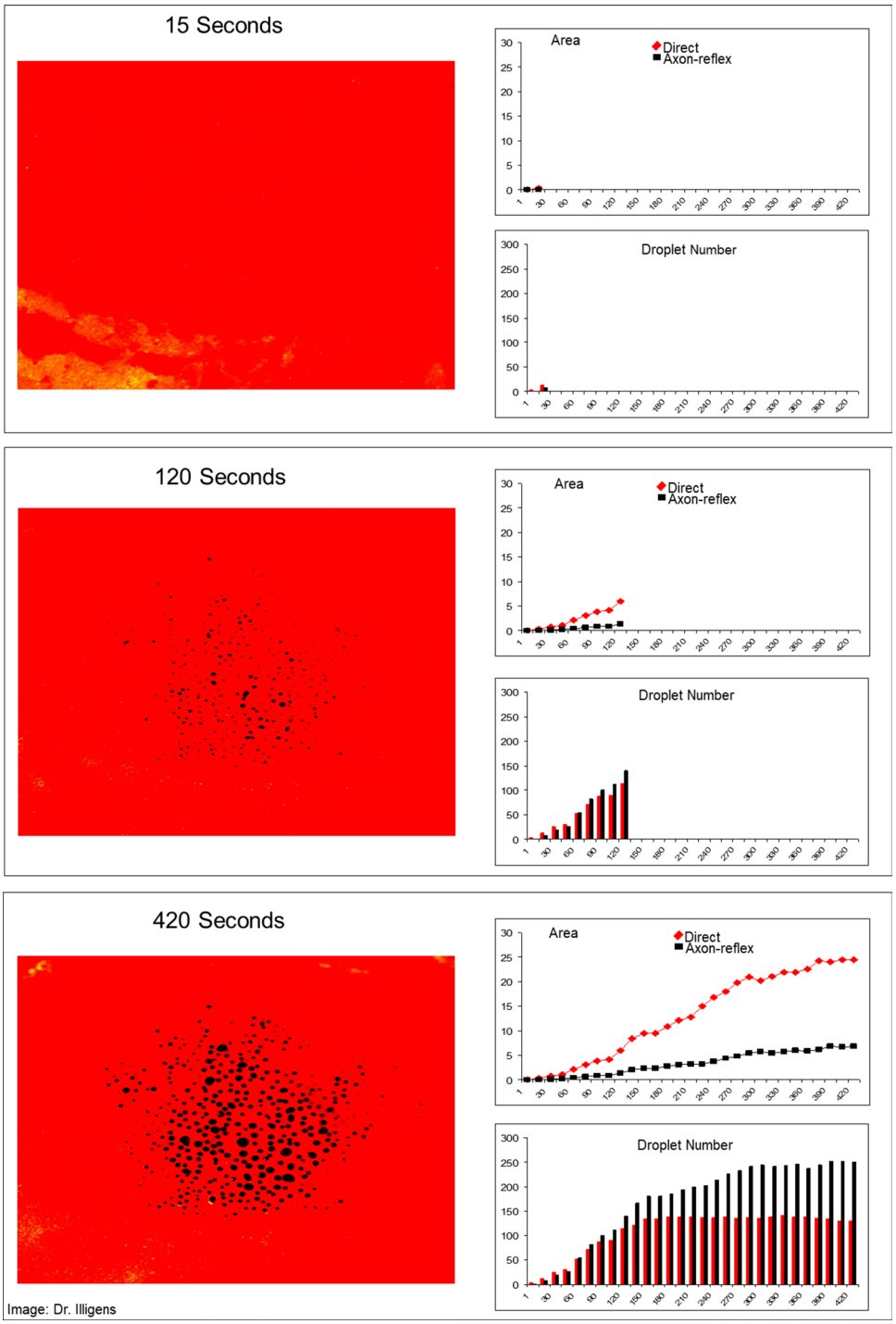

Fig. 4 QDIRT responses over time. Digitalized analysis of acetylcholine induced sweating with temporal and spatial resolution. Three different time points of sweat pattern analyses are displayed with photographs of the cutaneous sweat response on the left side and diagrams showing the axon-reflex spread area as well as the sweat droplet count on the right side

utilized the QDIRT technique [1]. Therefore, normative data are not available.

Perspective

Studies comparing QDIRT with the other sudomotor function testing procedures in patients affected by the different causes of autonomic peripheral neuropathies, e.g., diabetes or amyloidosis, are needed to evaluate the sensitivity and specificity of the QDIRT in a clinical setting. A longitudinal multicenter study of sudomotor and pilomotor function to assess progression of synucleinopathic small fiber neuropathy in patients with Parkinson's disease and healthy subject is currently under way. This 3-year protocol includes sudomotor assessment via QDIRT as well

as sympathetic skin response (SSR) and might add to the evaluation of the clinical utility of QDIRT [19].

Electrochemical skin conductance

Similar to SSR, electrochemical skin conductance (ESC) is based on the detection of quantitative changes of electrochemical carriers on the skin surface. ESC devices provide a non-invasive way to study sweat gland function and, hence, indirectly the sudomotor function [20, 21]. The patient is instructed to put both palms and both feet simultaneously on nickel electrodes, which are connected to a computer equipped with specialized analysis software. Additionally, a headband can be used to evaluate the sweat response on the forehead. Testing time takes approximately 2 min and is completely automatically. The measured ESC is stated in µSi [22]. High values of the ESC indicate a lower risk of sudomotor dysfunction [22].

Neurophysiological background

Measurement of ESC is based on both reverse iontophoresis and chronoamperometry measuring the chloride ion concentration [23]. A low direct-current incremental voltage is applied to the anode, inducing a voltage to the cathode electrode. The current initiates a shift of chloride ions from the sweat glands [20]. The resulting current between the electrodes is proportional to the chloride concentration on the skin surface [23]. However, to date it remains to be answered, whether the induced change of ESC stimulates sudomotor fibers or the sweat glands directly [24].

Clinical implications

Automatic measurement of ESC is relatively easy to conduct and can be performed outside of specialized autonomic centers. It allows an early detection of small nerve fiber damage, e.g., in patients with diabetic polyneuropathy. Particularly for diabetic patients, changes of ESC are well-studied and have shown to be valuable to predict the risk for autonomic cardiac failure [24]. Moreover, minor patient afford is an advantage for the clinical implementation of ESC measurement.

Limitations

One major limitation of ESC is the display of interindividual differences in eccrine sweat gland function. In general, ESC values display a sweat gland dysfunction, whereas differentiation of postganglionic neurological and non-neurological disorders as cause of sudomotor dysfunction is not possible [24]. Moreover, normative data are still under investigation, as studies have revealed, that ESC differs between different groups of patients, e.g., a decreased change in ESC in obese control groups and a dependency on ethnicity [20, 24]. In patients with pre-diabetes, type 2 diabetes or chronic kidney disease, a decrease of ESC was described [24]. In contrast, ESC was not reduced in patients with longstanding type 1 diabetes [24]. Interestingly, there are no gender differences of ESC reported, which might be considered as an advantage compared to the other available sudomotor function assessment tests [24].

Additionally, the mechanism of reversed iontophoresis might be less specific for the analysis of small nerve fiber function compared to stimulation of the axon-reflex. Particularly, the influence of certain medications or possible electrolyte adjournments on changes of ESC is unknown. This might be important since the drifting of chloride ions is the major diagnostic target of ESC changes. Finally, the presence of dermatological disorders could change the chloride ion drifting. Notable, the ESC measurement showed a high reproducibility in studies [24].

Perspective

ESC measurement might benefit from viewing the changes in conjunction with patients' blood electrolyte concentrations. The method might facilitate the screening of patients for autonomic peripheral neuropathies as well as their response to treatment. However, ESC measurement has not the potential to replace the gold-standard sudomotor function test yet [24]. Further studies on different devices and ESC changes in different patient groups (e.g., to further study the influence of BMI, age and ethnicity) to define normative data are necessary.

Sensitive sweat test

The SST has been introduced by Loavenbrock to evaluate the sweat secretion of each sweat gland along with their number, location and distribution [25]. In a pilot study, the postganglionic sudomotor function in healthy controls and subjects with diagnosed distal sensorimotor neuropathy was assessed by stimulating the sudomotor through iontophoresis of 0.5% pilocarpine solution followed by skin drying and staining of the stimulated area with 10% povidone-iodine solution [25]. The tested body sites in the control subjects have been: foot dorsum (distal to extensor digitorum brevis muscle), medial-posterior calf (one-third the distance between knee and ankle), distal medial thigh (over motor point of the vastus medialis muscle), and the dorsal hand (overlying first dorsal interosseous muscle) [25]. In this study the neuropathy patients have been tested at the foot and calf [25]. Afterwards, a miniature video camera (13 × 17.5 mm

field of view and 29 mm focal length) was placed on the iontophoresis area. The lens of the video camera is equipped with a transparent tape covered with a thin layer of corn starch [25]. Automatic video recording of the sweat initiated color change of the indicator was recorded. The area of each iodine–starch-spot is proportional to the volume of sweat produced by the underlying sweat gland. The enlargement rate of each spot is proportional to the sweat production rate of that spot's underlying sweat gland [25]. For evaluation of the sweat gland function an analysis software was utilized, which calculated pixel-based, after calibration with standard solutions, the sweat rate and volume. A second recording needs to be conducted after swapping a new starch tape on the camera. Therefore, the skin is fully covered with povidone-iodine solution and dried for replicate analysis.

Neurophysiological background

In contrast to QSART, silicone imprints and QDIRT, in the SST pilocarpine is delivered via iontophoresis into the upper dermal skin layers. Pilocarpine is a cholinergic agonist, directly stimulating the sweat glands by binding to tubular M3-receptors of the sweat glands. An axon-reflex mediated activation of nearby sweat glands is not described for pilocarpine. Fully denervated nerve fibers are non-responsive to pilocarpine stimulation, which is utilized to assess the function of each sweat gland. Maximal effects after pilocarpine injection are seen after 10 min and decreasing sweat response is seen 20 min post-injection [26].

Clinical implications

Being potentially useful in the investigation of single sweat gland function in a relatively fast and rather uncomplicated setting, the SST might complement other easy-to-perform techniques for the assessing autonomic peripheral neuropathies by the analyzation a temporal and spatial resolution of an evoked sweat response [25]. If further investigations endorse the capability of the SST for evaluation of sudomotor function, SST would make clinical sudomotor testing both for patients and performing clinical staff less burdensome [25].

Limitations

Further investigation of this new sudomotor function testing with larger study cohorts is needed to confirm the capability of the SST in routine autonomic nervous system testing. Moreover, the primarily published testing protocol is time consuming for both the patients and the conducting clinicians compared to, e.g., the QDIRT or QSART. Training of clinical staff seems to be essential to achieve the highest possible accuracy. Possible influencing environmental factors need to be considered. While SST assesses sweat gland function, it lacks the possibility to evaluate nerve fiber function by studying axon-reflex responsiveness. Moreover, as sudomotor function tests based on indicator dye reactions, the quantification of sweat reaction over time is aggravated by proceeding sweat droplet cohesiveness, which allows only a short-term analyzation of individual sweat secretion.

Perspective

Studies with larger sample sizes are needed to show the reliability and reproducibility of the SST and to define normative values. As mentioned by Loevenbruck et al., the use of acetylcholine or other axon-reflex mediating substances could help to better understand the physiology of the sudomotor function and allow evaluation of axon-reflex responsiveness [25].

Sympathetic skin response

The SSR technique assesses electrodermal activity upon stimulation of sudomotor nerves. In 1984 Shahani and colleagues emphasized that SSR might provide a simple way to test unmyelinated axon function in peripheral neuropathies, introducing the further use of this method [27]. To conduct a SSR, a surface electromyography electrode is placed on the patient's palm or sole accompanied by a reference electrode on each dorsal side of the tested body area. Most commonly the change of the skin potential is evoked with an electrical stimulation, less often by breathing maneuvers (deep inspiration). The frequency filters of the recording computer, more specifically of the software, are set low (usually to 0.1–0.5 Hz) due to the low increase of the electrodermal potential caused by the sweat appearance on the skin surface [28]. To achieve accurate SSR recordings, the tested skin area should be well prepared prior to SSR. The testing room should be light dimmed, and the room temperature should be kept between 22–24 °C to avoid any confounding. Humidity control is also important to reduce environmental confounding. The recorded SSR graphs are analyzed for presence/absence and for latency and amplitude of the electrical potential change. A missing SSR may require a second electrical stimulation or, in case of a breathing evoked sweat response, further instruction of the patient. Usually, the palmar SSR has a shorter latency, but higher amplitude than the plantar recorded SSR (hands 1.5 s latency, 0.5–1.3 mV amplitude, feet 1.9–2.1 s latency, 0.15–0.8 mV amplitude) [29].

Neurophysiological background

The SSR reposes on a change of the skin potential after electrical stimulation or deep inspiration of the patient. The electrodermal activity is mainly influenced by the secretory eccrine sweat glands, as sweat consists of mainly water and small amounts of electrolytes, e.g., sodium and potassium, controlled by spinal, bulbar and suprabulbar centers [1, 28]. However, the SSR as somato-sympathetic reflex is not fully understand yet.

Clinical implications

Sudomotor function is easily accessible with SSR and allows a precise examination of sudomotor function, if SSR is conducted equitable to a standard protocol. Moreover, SSR has been investigated in multiple studies in patients with spinal cord injuries and has been shown to be a helpful diagnostical test to determinate both the spinal sympathetic function and the peripheral sympathetic function [30]. Due to its high sensitivity toward changes in electrodermal activity due to emotional response, SSR is also used to record the emotional sweat response in the form of lie detectors and psychophysiological studies [31].

Limitations

Absent SSR after stimulation can occur due to habitation. Moreover, it has been reported that patients aged 50 years or more tend to have no recordable SSR [29]. A large interindividual variability in SSR has been described, thus SSR interpretation should be performed together with other sudomotor function tests [25]. While SSR results are helpful to compare sudomotor function between groups of individuals, their high intersubject variability limits its use in individual patients [29]. Due to this variability, reliable normative data on latency and amplitude of the response are not available. However, complete absence of SSR potentials, particularly if one-sided, can be helpful in diagnosing neurological conditions when viewed in conjunction with other neurophysiological tests such as sensory and motor electroneurography [25]. Additionally, it should be noted that patients with ectodermal anhidrotic dysplasia (congenital absence of sweat glands) still have a sympathetic skin response. Due to limited sensitivity and specificity of the technique, SSR as a single method is only a surrogate measure of sudomotor function [28].

Perspective

Although a variety of studies have characterized sensitivity and specificity of the SSR, the underlying central mechanism of this somato-sympathetic reflex remains relatively unknown until today [25]. Therefore, further neurophysiological and neuroanatomical basic research remainsin order to fully understand the SSR [32].

Spoon test

The spoon test was first published by Bors as a bedside test to access sudomotor function in patients with autonomic failure [33]. This non-quantitative sudomotor function screening test is based on the phenomenon of the interruption of smooth sliding of the convex side of a spoon on dry skin [33, 34]. It has been recently compared to other screening instruments of the sudomotor function [35]. In a method comparison study, the spoon test demonstrated the highest specificity and sensitivity when performed over the skin of the chest and forehead. This observation indicated that the spoon conducted test might be a valid bedside screening instrument in these specific skin areas.

Neurophysiological background

The spoon test qualitatively assesses the smoothness sliding of the convex side of spoon on moist skin. Therefore, it does not require any pharmacological stimulation of the sweat glands.

Clinical implications

The spoon test can be used as a pre-TST bedside sudomotor screening instrument. It requires less trained staff, is inexpensive and has shown high sensitivity and specificity in defined skin areas [35]. However, the technique lacks any quantitative analysis and results depend on the tester's sensation for the spoon sliding. A recent study revealed the spoon test as more sensitive and specific as other available qualitative bedside screening tools, such as visual skin inspection [35]. However, it should be highlighted, that the spoon test should only be used as a basic screening tool of sudomotor function.

Limitations

The spoon test does not allow a quantification of sudomotor function and depends on the tester's sensation for the spoon sliding. Therefore, the spoon test is a non-objective screening test of sudomotor function, which consequently allows only a schematic screening of patients' sudomotor function. However, it might improve the bedside detection of sudomotor dysfunction in patients with clinical sudomotor dysfunction symptoms by physicians, that are not trained in autonomic nervous system assessment. This might result in an improved screening of sudomotor dysfunction and

Table 1 Overview of sudomotor function tests

	TST	QSART	Silicone imprint	QDIRT	Sudoscan	SST	SSR	Spoon test
Detected sudomotor alteration	Pre- and postganglionic	Postganglionic; axon reflex	Pre- and postganglionic	Postganglionic; axon reflex	Unclear	Postganglionic	Pre- and postganglionic	Pre- and postganglionic
Tested body site	Ventral body surface	Forearm, proximal and distal leg; dorsal foot	Distal limbs	Distal limbs	Palmar and plantar, forehead	Healthy subjects: foot dorsum, medial-posterior calf, distal medial thigh, dorsal hand neuropathy patients: foot dorsum, medial-posterior calf	Palmar, plantar	Total body surface
Reproducibility	Good	Good	Good *(low-thresholded screening tool)*	Good	High *(automatic testing and analyzation)*	Good	Low, high intra-individual variability, habitation	Good *(low-thresholded screening tool)*
Normative data	Symmetric sweat pattern over ventral body surface	Sweat response latency: ~1–2 min maximum sweat response: ~5 min mean sweat output females = 0.25–1.2 µl/cm^2 males = 2–3 µl/cm^2	Hand: 311 ± 38 sweat droplets/cm^2 *(lower limit 255 droplets)* foot: 281 ± 38 sweat droplets/cm^2 *(lower limit 235 droplets)*	Under current investigation	Under current investigation	Under current investigation	Hand: 1.5 s latency; 0.5–1.3 mV amplitude; foot: 1.9–2.1 s latency; 0.15–0.8 mV amplitude)	No quantification
Confounding variables[a]	Sweat droplet cohesiveness; influence of indicator dye reaction	Long-established preganglionic dysfunction; early stage small-fiber neuropathy; reflex sympathetic dystrophy	Congenital lack of eccrine sweat glands; silicone related inaccuracies	Sweat droplet cohesiveness; influence of indicator dye	Differences in eccrine sweat gland function	Sweat droplet cohesiveness	Age, differences in eccrine sweat gland function, sweat composition	Depending on tester's sensation

[a]Known confounding variables for all sudomotor function tests: patients' body temperature, nicotine, caffeine and hydration status; humidity and room temperature

increasing number of referrals to specialized autonomic centers for specific assessment and treatment.

Clinical staff needs to be trained to develop a sense for the sweat-triggered interruption for smooth gliding of the spoon. Standardizing of this test is difficult as the way the spoon is moved over the skin surface depends on the examiner's arm movement. Moreover, further evaluation of the spoon test with respect of specificity and sensitivity in larger cohorts and under different testing conditions remain needed.

Perspective

A future experimental development could be the connection of a speedometer to the spoon, which could help to gather quantitative data about the sudomotor function. However, this would require standardization of the applied pressure as well and thereby compromising feasibility of the technique as an easy-to-perform bedside test [35].

Conclusion

There are several available methods to evaluate the sudomotor function, which allow the evaluation of patients reporting symptoms indicative of sudomotor dysfunction such as impaired heat tolerance or anhidrotic skin areas (Table 1). Over the last decades sweat testing has become an essential component of autonomic nervous system testing, explainable by the fact that sudomotor function is among the earliest signs of a variety of small fiber neuropathies including diabetic and toxic small fiber neuropathy [3, 12]. Only a few of the described tests have been shown feasible outside non-specialized centers such as the SSR in patients with neuropathiesy [3]. Their clinical use is, however, limited by substantial between-subject variability. More precise techniques such as QSART and TST are limited by high technical and staff-related demands. However, research has recently focused on simplifying testing protocols to allow broader use of sudomotor assessment. Consequently, techniques such as QDIRT and measurement of changes of ESC have been introduced. While QDIRT might improve precision of previous postganglionic small fiber tests, the technique is limited by a substantial lack of data on its use in pathological conditions [3]. By contrast, measurement of ESC has been tested in several studies but the association of assessed changes of electrolyte movement upon reversed iontophoresis and sudomotor nerve function are not fully elucidated, limiting interpretability of measurement results [3]. The spoon test allows only a bedside screening for sudomotor dysfunction and should not be used as a clinical tool to fully examine sudomotor function.

In specialized autonomic nervous system laboratories, a combination of TST and QSART has been established as a gold standard in the general autonomic testing battery neuropathy [3]. The TST acts as a screening tool to study the sudomotor function of the ventral body surface. The additional conduction of QSART allows differentiating between pre- or postganglionic origin of sudomotor dysfunction. Since the autonomic nervous system is influenced by multiple factors including avoidable influencing factors, such as intake of sympathomimetic, anticholinergic or carbonic anhydrase-inhibiting medications, these drugs should be stopped, if possible, for at least 24–48 h prior to sudomotor function testing (depending on their specific pharmacological half-life).

However, in a clinical setting, testing results need to be interpreted in conjunction with individual medical history and symptoms as well as findings of additional diagnostic tests such as neuroimaging of autonomic centers and peripheral electroneurography of somatic nerves.

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Compliance with ethical standards

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