Concordance between sudomotor disorder and the clinical diagnosis of diabetic peripheral neuropathy, according to various clinical guidelines

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ABSTRACT:

Aim. To assess the concordance between variations in Neuropad® results and the those in different diagnostic criteria of Diabetic Peripheral Neuropathy, according to various clinical guidelines**. Methods:** A descriptive observational study was conducted of 111 patients with a confirmed diagnosis of Diabetes Mellitus. The criteria for inclusion in the study were that patients should be aged 18 years or more and have at least 10 years' history of Diabetes Mellitus. **Results:** 73 (65.8%) were male and 38 (34.2%) were female. Their mean age was 57.92 ± 13.24 years (95% CI 55.45-60.38). Healthy Neuropad® findings were obtained for 35 right feet (31.5%) and 31 left feet (27.9%). **Conclusion:** Neuropad® is an effective instrument for detecting macro and microvascular complications such as early-stage neuropathy, although its use should always be accompanied by a clinical examination of the foot.

KEY-WORDS

Clinical Guidelines; Diabetic Peripheral Neuropathy; Diabetes Mellitus; Neuropad.

1. INTRODUCTION

In 2017, according to the International Diabetes Federation, 425 million people worldwide, or 8.8% of all adults aged 20-79 years, were living with diabetes mellitus (DM), about four times more than in 1980. Regrettably, this figure is expected to increase to about 642 million by 2040 [1]. DM is the seventh leading cause of death and provokes many debilitating, detrimental consequences, including organ failure, heart disease, stroke, blindness and lower-extremity amputations [2].

One in seven patients with diabetes will develop some type of foot injury during their life. Among the most feared and common complications of diabetes are ulcers of the foot [3]. The lifetime incidence of diabetic foot ulceration is 19-34%, and the yearly incidence is 2% [4]. After successful healing, the recurrence rates of diabetic foot ulcers (DFU) are 40% within a year and 65% within 3 years. Therefore, in order to reduce risks to the patient and the resultant economic burden to society it is of paramount importance to prevent DFU. Not all patients with diabetes are at risk of ulceration; major risk factors include a loss of protective sensation, peripheral artery disease and foot deformity. A history of foot ulceration and any level of lower extremity amputation further increase the risk of ulceration [5]. In general, patients not presenting any of these risk factors do not appear to be at risk of ulceration. Loss of protective sensation or diabetic peripheral neuropathy (DPN) is a common complication of diabetes, affecting more than 50% of patients with diabetes aged over 60 years[6].

Sudomotor dysfunction is a characteristic feature of DPN. The sweat glands are innervated by long, thin, unmyelinated fibres of the sympathetic nervous system, which are damaged in the early stages of diabetes, leading to hyposudoration or anhydrosis in the lower extremities, with the consequent appearance of cracks in the feet that may facilitate the entry of infection [7]. The

American Diabetes Association and the Toronto Research Group both recommend assessing the skin for this condition [8][9]. It is vitally important, therefore, to identify the presence of DPN in order to stratify the risk presented in this respect to patients with diabetes[10]. This diagnosis is normally obtained by the examination of diverse sensory modalities using simple clinical tests[11]. In daily practice, however, the diagnosis of neuropathy with existing tests is often suboptimal, especially when the condition is incipient and affects the small fibres. Tests that have been developed to improve the diagnostic yield [12] in the detection of somatic and, to a lesser extent, autonomic neuropathy include Neuropad®[13–17] and SudosCan[18][19].

Neuropad® has been compared with other tools used in the screening of patients with diabetes, such as the Neuropathy Disability Score, the Michigan Neuropathy Screening Instrument-Questionnaire, the 10-g monofilament test and the diabetic neuropathy test. Neuropad® detects small-fibre damage, which is not detected by most other tests, but to our knowledge no data have been reported concerning its ability to detect early diabetic neuropathy[20].

The aim of this study is to assess the concordance between variations in Neuropad® results and the those in different diagnostic criteria of DPN, according to various clinical guidelines.

2. MATERIAL AND METHODS

2.1. Participants: A convenience sample was obtained from 111 patients with a confirmed diagnosis of DM. As design a descriptive observational study was carried out. The criteria for inclusion in the study were that patients should be aged 18 years or more and have at least 10 years' history of DM. Patients with distal foot amputation and the presence of significant hyperkeratosis in the forefoot area, preventing the placement of the Neuropad®, were excluded.

2.2.Ethical approval: This study was approved by the ethics committee at the Hospital (Reg. No. 13/305) and was performed in accordance with the Declaration of Helsinki and all applicable legislation and regulations on ethical standards for human experimentation. Signed informed consent was obtained from all participants.

2.3.Methods

Two researchers independently interviewed the patients to obtain the study data. The clinical interview was conducted in one room, where the patients were asked to complete a questionnaire about sociodemographic variables, the type of diabetes and its duration, the presence of toxic habits (smoking) and the frequency of physical activity. Subsequently, the patient's weight, height and brachial blood pressure were determined.

Another investigator then performed the DPN examination of the feet, after which the Neuropad® was attached to the plantar area (Anexe I).

2.4.Data collection

The degree of metabolic control was determined according to the last analysis performed in the previous six months (blood glucose, glycated haemoglobin (HbA1c), triglycerides, total cholesterol, High-Density lipoproteins (HDL) cholesterol, Low-Density Lipoproteins (LDL) cholesterol, creatinine). The following comorbidities were recorded: high blood pressure, coronary heart disease, cerebrovascular diseases, nephropathy, retinopathy and/or cancer. To detect the presence of the risk factor of smoking, patients were asked if they were or had been smokers.

Following the recommendations of the American Heart Association [21], blood pressure was measured in the arm considered dominant (the one with the highest systolic blood pressure),

using a correctly-calibrated Colin BP-8800C automatic measuring device. The Body Mass Index (BMI) was determined by asking the patient to stand barefoot on the Tanita TBF 300 calibrated scale. Height was measured using a Harpenden® stadiometer.

The possible presence of peripheral artery disease was determined by calculating the ankle-brachial index with a bidirectional Smartdop 45 Hadeco® Doppler probe.

The presence/absence of DPN was determined according to five criteria stipulated in various clinical guidelines (ADA 2018, IWGDF 2016, IDF 2012)[10]. The clinical examination was conducted by applying a 5.07 Sensifil™ monofilament (Novolab Iberica®) perpendicular to the skin. Vibratory sensitivity was assessed on a scale ranging from 0 to 8, using a 128 Hz Rydel-Seiffer tuning fork. Sensitivity to pain was explored using the Neuropen®, an instrument that presents a blunt non-puncturing tip attached to a base (lancet), with a spring that exerts a pressure of 40 gr, sufficient to provoke a painful stimulus. The Achilles reflex was examined by tapping the Achilles tendon with a rubber hammer, with the knee flexed and resting on a surface, to determine the presence of plantar flexion in the foot.

Finally, sudomotor dysfunction was assessed by the application of Neuropad®, an adhesive patch containing cobalt chloride anhydride salt, which is placed on the skin to detect any abnormality in the sympathetic nervous system. This patch was placed in the plantar area of the foot, between the first and second metatarsophalangeal joints, avoiding areas of hyperkeratosis [14] The test was performed indoors, at a temperature of 20-25 °C, for ten minutes, after which the colour change was assessed. The patient's condition is considered normal when the colour of the Neuropad® changes from blue to pink.

2.5.Statistical analysis

The statistical analysis was performed using SPSS version 26.0 for Windows (SPSS Inc, Chicago, USA). The Student t test for independent samples was performed to compare mean values of the quantitative variables. The v2 statistical test was used to identify differences in the proportions of qualitative variables. Odds ratios and the corresponding 95% confidence intervals were determined by univariate and multivariate logistic regression models. A 5% type I error rate was assumed (p= 0.05).

3. RESULTS

Although 153 persons were initially analysed, only 111 fully met the criteria for inclusion. Of these, 73 (65.8%) were male and 38 (34.2%) were female. Their mean age was 57.92 ± 13.24 years (95% CI 55.45-60.38) and the average duration of their DM was 17.59 ± 10.70 years (95% CI 15.59-19.59). Of the patients included, 67 (60.9%) did not have good metabolic control (HbA1c >7%) and 53 (47.7%) exceeded the threshold of obesity (BMI >30 Kg / m^2) (see Table 1).

The patients with abnormal Neuropad® results had a higher mean age (60.33 ± 11.94 years) than those whose results in this respect were normal (51.10 ± 14.54); p = 0.004 (95% CI 3.80-14.64). The other sociodemographic and lifestyle variables (tobacco, daily exercise) were not statistically significant.

A significantly greater association was observed between abnormal Neuropad® results and a history of hypertension (p = 0.002; OR 3.81; 95% CI 1.56-9.27), retinopathy (p = 0.002, OR 4.80, 95% CI 1.66-13.83) and coronary heart disease (p = 0.003, OR 1.46, 95% CI 1.27-1.68) than among patients with no such comorbidities.

Furthermore, the patients with abnormal Neuropad® results had significantly lower HDL values (50.33 ± 15.84) than those whose Neuropad® results were within normal limits (57.79 ± 17.7, p = 0.04. Creatinine values were also significantly higher among the patients with altered Neuropad® (0.94 ± 0.29).

Healthy Neuropad® findings were obtained for 35 right feet (31.5%) and 31 left feet (27.9%). The test results were pathological (i.e., blue or with blue traces) in 76 right feet (68.5%) and 80 left feet (72.1%) (see Table 2). Altered Neuropad® values were obtained in one or both feet for 82 patients (73.9%).

Altered Neuropad® values were consistent with a diagnosis of DPN obtained by the monofilament test alone in four areas of the foot (right foot p = 0.029 Sensitivity (SE) 0.88, Specificity (SP) 0.37; likelihood ratio positive (LK+)1.40; left foot p = 0.044 SE 0.89, SP 0.33; LK+ 1.33); with the monofilament + pinprick test in the left foot (p = 0.057; SE: 1.00, SP 0.32; LR+ 1.47); with the monofilament test + tuning fork in the right foot (0 = 0.032; SE 0.9, SP 0.37; LR+ 1.43) and with the monofilament test + cotton wisp in the left foot (p = 0.013; SE 1.00; SP 0.32; LR+ 1.48) (see table 3)

4. DISCUSSION

The aim of this study is to determine the level of concordance between an assessment of DPN based on altered Neuropad® values and that derived by applying different diagnostic criteria, according to the respective clinical guidelines.

Our results show that 54% of patients with no clinical diagnosis of DPN according to the monofilament sensitivity test nevertheless presented abnormal Neuropad® values, indicative

of motor involvement in the early stages of a neuropathy. This suggests that the Neuropad® test could be a valuable means of early detection of DPN, as has been observed in previous studies [13,22–24]. Furthermore, Neuropad® is a diagnostic tool with high inter-user reproducibility [25]. Sudomotor dysfunction in patients with DM is associated with the presence of dry feet and elevated skin temperature, which are risk factors for the development of plantar ulcers and for possible amputation [26–28].

Greater body volume is expected to stimulate the activity of the sweat glands, thereby decreasing skin temperature and reducing the risk of ulceration. However, in our study, although the BMI was not a significant factor, it was higher (30.11 ± 5.39) in the patients with altered Neuropad® values, who presented less perspiration in the feet ($p = 0.781$). Another relevant factor is that increased age is associated with changes in the number of sweat glands identified by histological analysis [29], which could explain why our study revealed a significant relationship between age and altered Neuropad® values ($p = 0.004$, 95% CI 3.80-14.64).

Corroborating previous research, it was found the presence of sudomotor involvement to be associated with vascular complications such as hypertension, coronary heart disease [30] and microvascular complications [31] such as retinopathy [32]. However, unlike some other studies [33][34], it was found no significant relationship with the presence of nephropathy.

By correlating the different diagnoses of DPN according to the corresponding clinical guidelines, it was found that there was consistency between altered Neuropad® values and the diagnosis of DPN in both feet, using only the monofilament in four areas of the foot. This contrasted with Papanas [35], who observed correlation with the vibration perception threshold measured with a neurothesiometer. In another study, Didangelos et al.[36] showed

that Neuropad® has high sensitivity but only moderate specificity vs. the monofilament test in patients with DM. Another recent study reported that if the two tests (monofilament and Neuropad®) are used jointly as screening tools for patients with DM, plantar ulcers are less likely to appear and the cost to healthcare systems is reduced [37].

Among the strengths of this research, the diagnostic tests and the interviews with the patients were carried out at a constant room temperature of 20-25ºC with a ten-minute acclimatisation period followed by the test proper (duration: 10-15 minutes) and assessment of the colour changes observed. These conditions ensured that the patients were in a comfortable environment and not subject to any stress that might impact on the activity of the sweat glands. Furthermore, the colour change in the Neuropad® was compared with the results of the well-established monofilament test to identify early stages of DPN [10].

On the other hand, the study also presents important limitations. In particular, the sample size might be considered inadequate, the percentage of male patients significantly exceeded that of females, and the sample contained no patients of advanced age.

From the findings obtained, it conclude that the Neuropad® is an effective instrument for detecting macro and microvascular complications such as early-stage neuropathy, although its use should always be accompanied by a clinical examination of the foot. Moreover, it is suitable for self-testing. By enabling patients with diabetes to monitor their foot health at home, the Neuropad® provides a useful, economical means of freeing up time and other resources in the surgery and elsewhere in the health system.

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6. TABLES

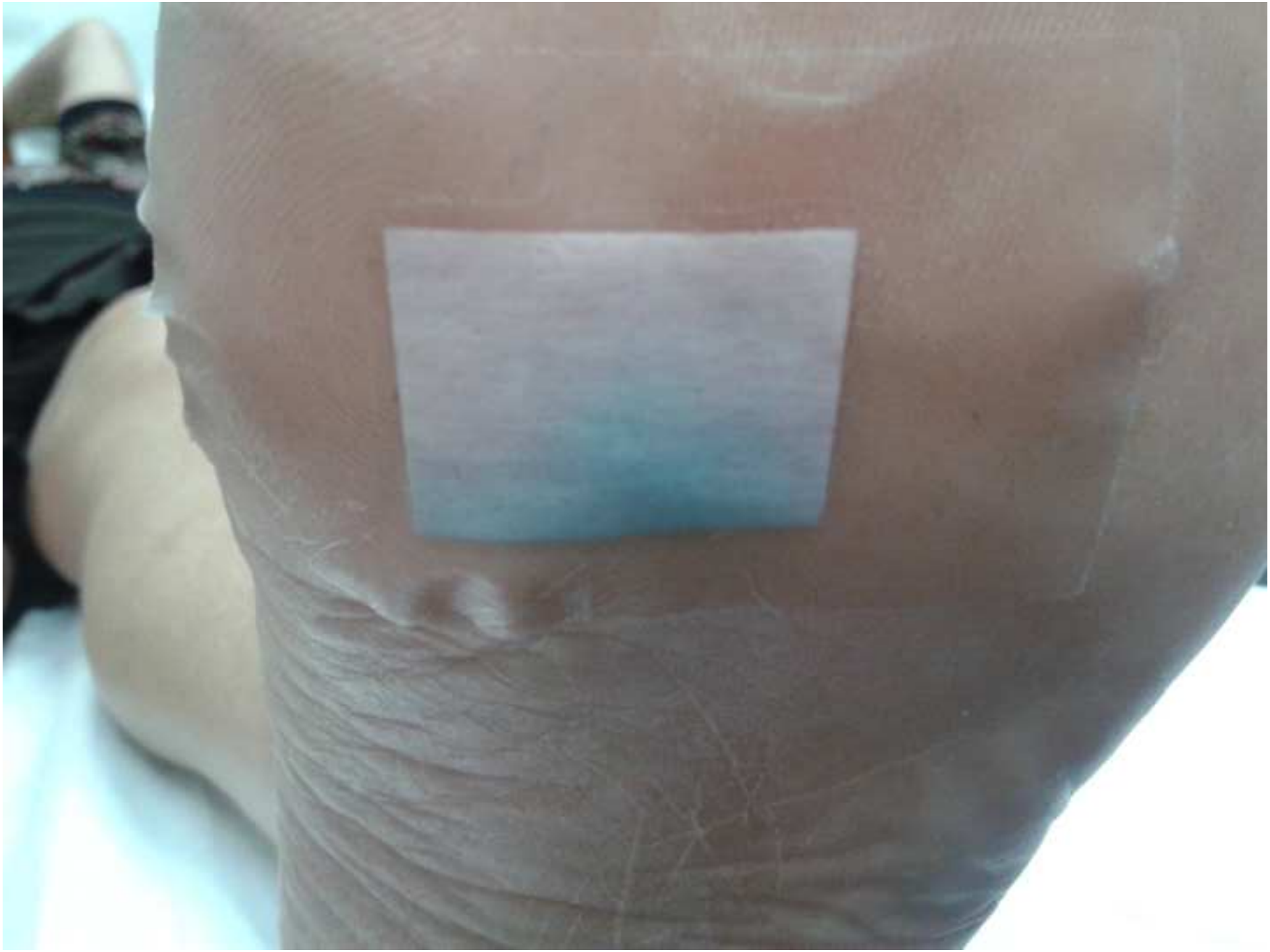

Table 1. Sociodemographic characteristics, lifestyle and clinical variables

Characteristics	Altered Neuropad® N=82	Correct Neuropad® N=29	p value
Sex (n [%]) Male/Female	55 (67.1)/27 (32.9)	18 (62.1)/11 (37.9)	p=0.625
Mean age ± SD [years] (95% CI)	60.33±11,94	51.10±14.54	**p=0.004** (95% CI 3.80-14.64)
Mean duration of diabetes ± SD [years]	18.23±11.15	15.73±9.18	p=0.288
DM type 1/ type 2 (n [%])	22 (26.8)/60 (73.2)	12 (41.4)/17 (58.6)	p=0.144
BMI >30 Kg/m² (n [%])	30.11±5.39	29.77±6.06	p=0.781
Mean SAP ± SD [mm Hg]	138.67±15.87	140.62±19.66	p=0.633
Mean DAP ± SD [mm Hg]	72.51±9.41	76.79±12.74	p=0.106
Peripheral arteriopathy (n [%])	31(38.3)	8 (27.6)	p=0.302
Hypertension (n [%])	62 (75.6)	13 (44,8)	**p=0.002** OR 3.81 (95% CI 1.56-9.27)
Nephropathy (n [%])	14 (17.3)	1 (3.4)	p=0.062
Retinopathy (n [%])	41 (50)	5 (17.2)	**p=0.002** OR 4.80 (95% CI 1.66-13.83)
Stroke (n [%])	5 (6.1)	1 (3.4)	p=0.588
Coronary heart disease (n [%])	20 (24.4)	0 (0%)	**p=0.003** OR 1.46 (95% CI 1.27-1.68)
Cancer (n [%])	7 (8.5)	1 (3.4)	p=0.362
Walking daily (n [%])	40 (48.8)	10 (34.5)	p=0.183
Smoker (n [%])	52 (63,4)	17 (58.6)	p=0.647
Mean HbA_1c ± SD	7.41±1.20	7.34±0.95	p=0.793
Triglycerides ± SD [mg/dl]	116.80±62.20	98.54±56.78	p=0.175
Mean cholesterol LDL ± SD [mg/dl]	93.49±27.73	86.11±26.26	p=0.222
Mean cholesterol HDL ± SD [mg/dl]	50.33±15.84	57.79±17.72	**p=0.040** (CI95% -14.65/-0.34)
Creatine ± SD [mg/dl]	0.94±0.29	0.82±0.17	**p=0.048** (95% CI 0.001-0.23)

DM, Diabetes mellitus, BMI, Body mass index (calculated as weight in kg divided by height, in m, squared); SAP, Systolic blood pressure; DAP, Diastolic blood pressure; HbA_1c, Glycated haemoglobin; LDL, Low density lipoprotein; HDL, High density lipoprotein. SD, standard deviation; OR: Odds ratio; CI, Confidence interval.

Table 2. Neuropad® results

Neuropad® colour change	n=111	
	Right foot n (%)	Left foot n (%)
Neuropad® colour: pink (n [%]	35 (31.5)	31 (27.9)
Neuropad® colour: almost pink (doubtful) (n [%])	13 (11.7)	15 (13.5)
Neuropad® colour: blue traces (n [%])	58 (52.2)	58 (52.2)
Neuropad® colour: blue (n [%])	5 (4.5)	7 (6.3)

Table 3. Neuropad® results for different diagnostic criteria of neuropathy

NEUROPAD® COLOUR CHANGE	Monofilament (4 areas)				Monofilament (4 areas) + Pinprick				Monofilament (4 areas) + tuning fork				Monofilament + Achilles reflex				Monofilament + cotton wisp			
	No DPN		Presence of DPN		No DPN		Presence of DPN		No DPN		Presence of DPN		No DPN		Presence of DPN		No DPN		Presence of DPN	
	Right n (%)	Left n (%)	Right n (%)	Left n (%)	Right n (%)	Left n (%)	Right n (%)	Left n (%)	Right n (%)	Left n (%)	Right n (%)	Left n (%)	Right n (%)	Left n (%)	Right n (%)	Left n (%)	Right n (%)	Left n (%)	Right n (%)	Left n (%)
Neuropad® colour: blue (n [%])	3 (3.5)	3 (3.6)	2 (8)	4 (14.8)	4 (4.2)	4 (4.2)	1 (7.7)	2 (16.7)	3 (3.3)	5 (5.5)	2 (9.5)	2 (10)	3 (3.4)	3 (3.7)	1 (5.3)	3 (12.5)	4 (4.3)	4 (4.2)	1 (5.9)	3 (21.4)
Neuropad® colour: blue traces (n [%])	44 (51.2)	42 (50)	14 (56)	16 (59.3)	46 (48.4)	49 (50.5)	9 (69.2)	8 (66.7)	46 (51.1)	45 (49.5)	12 (57.1)	13 (65)	43 (49.4)	41 (50)	13 (68.4)	16 (66.7)	45 (48.9)	48 (50)	11 (64.7)	9 (64.3)
Neuropad® colour: almost pink (n [%])	7 (8.1)	11 (13.1)	6 (24)	4(14.8)	12 (12.6)	13 (13.4)	1 (7.7)	2 (16.7)	8 (8.9)	12 (13.2)	5 (23.8)	3 (15)	10 (11.5)	10 (12.2)	2 (10.)	2 (8.3)	10 (10.9)	13 (13.5)	3 (17.6)	2 (14.3)
Neuropad® colour: pink (n [%]	32 (37.2)	28 (33.3)	3 (12)	3 (11.1)	33 (34.7)	31 (32)	2 (15.4)	0 (0)	33 (36.7)	29 (31.9)	2 (95)	2 (10)	31 (35.6)	28 (34.1)	3 (15.8)	3 (12.5)	33 (35.9)	31 (32.3)	2 (11.8)	0 (0)
P value	**P=0.029** SE 0.88 SP 0.37 LR+ 1.40	**P=0.044** SE 0.89 SP 0.33 LR+ 1.33	**P=0.029** SE 0.88 SP 0.37 LR+ 1.40	**P=0.044** SE 0.89 SP 0.33 LR+ 1.33	P=0.419 SE 0.85 SP 0.35 LR+ 1.30	**P=0.057** SE 1.00 SP 0.32 LR+ 1.47	P=0.419 SE 0.85 SP 0.35 LR+ 1.30	**P=0.057** SE 1.00 SP 0.32 LR+ 1.47	**P=0.032** SE 0.90 SP 0.37 LR+ 1.43	P=0.247 SE 0.90 SP 0.32 LR+ 1.32	**P=0.032** SE 0.90 SP 0.37 LR+1.43	P=0.247 SE 0.90 SP 0.32 LR+ 1.32	P=0.372 SE 0.84 SP 0.32 LR+ 1.32	P=0.081 SE 0.88 SP 0.34 LR+ 1.33	P=0.372 SE 0.84 SP 0.32 LR+ 1.32	P=0.081 SE 0.88 SP 0.34 LR+ 1.33	P=0.270 SE 0.88 SP 0.36 LR+ 1.38	**P=0.013** SE 1.00 SP 0.32 LR+ 1.48	P=0.270 SE 0.88 SP 0.36 LR+ 1.38	**P=0.013** SE 1.00 SE 0.32 LR+ 1.48

SE: Sensitivity; SP: Specificity; LR+: likelihood ratio positive