Continuous subcutaneous insulin infusion preserves axonal function in type 1 diabetes mellitus†

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Abstract

Background Diabetic peripheral neuropathy is a common and debilitating complication of diabetes mellitus. Although strict glycaemic control may reduce the risk of developing diabetic peripheral neuropathy, the neurological benefits of different insulin regimens remain relatively unknown.

Methods In the present study, 55 consecutive patients with type 1 diabetes mellitus underwent clinical neurological assessment. Subsequently, 41 non-neuropathic patients, 24 of whom were receiving multiple daily insulin injections (MDII) and 17 receiving continuous subcutaneous insulin infusion (CSII), underwent nerve excitability testing, a technique that assesses axonal ion channel function and membrane potential in human nerves. Treatment groups were matched for glycaemic control, body mass index, disease duration and gender. Neurophysiological parameters were compared between treatment groups and those taken from age and sex-matched normal controls.

Results Prominent differences in axonal function were noted between MDII-treated and CSII-treated patients. Specifically, MDII patients manifested prominent abnormalities when compared with normal controls in threshold electrotonus (TE) parameters including depolarizing TE (10-20 ms), undershoot and hyperpolarizing TE (90–100 ms) (P < 0.05). Additionally, recovery cycle parameters superexcitability and subexcitability were also abnormal (P < 0.05). In contrast, axonal function in CSII-treated patients was within normal limits when compared with age-matched controls. The differences between the groups were noted in cross-sectional analysis and remained at longitudinal follow-up.

Conclusions Axonal function in type 1 diabetes is maintained within normal limits in patients treated with continuous subcutaneous insulin infusion and not with multiple daily insulin injections. This raises the possibility that CSII therapy may have neuroprotective potential in patients with type 1 diabetes. Copyright © 2014 John Wiley & Sons, Ltd.

Keywords type 1 diabetes mellitus; neuropathy; neuroprotective; CSII; MDI; insulin

Introduction

Diabetic peripheral neuropathy is a highly prevalent complication of diabetes mellitus (DM) affecting 30–50% of patients with long-standing disease [1]. This form of neuropathy typically manifests as a distal symmetric polyneuropathy, with patients presenting initially with sensory symptoms such as paresthesia and burning and later with motor symptoms such as weakness and wasting. Investigations of nerve pathology in diabetic patients have utilized a number of different techniques including quantitative sensory testing, nerve conduction studies and intraepidermal nerve fibre density [2–4]. These studies have found
reduced nerve fibre length and density and slowed conduction velocities. Animal models have suggested that changes in axonal properties may be due to altered function of axonal voltage-gated ion channels [5–12], mediated by hyperglycaemia [7,13,14].

When compared with type 2 diabetes, patients with type 1 diabetes (T1D) may develop a more severe phenotype, possibly mediated by insulinopenia and c-peptide deficiency, which are required for optimal axonal metabolism and function [14,15]. Recent studies utilizing nerve excitability techniques have suggested more prominent abnormalities in axonal function in patients with T1D compared with patients with type 2 diabetes in the absence of neuropathy [16].

Nerve excitability techniques provide information regarding the behaviour of various axonal ion channels, pump and exchangers that are involved in impulse conduction, information that cannot be gained from standard nerve conduction studies [17]. Studies in diabetic patients have demonstrated prominent changes in axonal ion channel function in both type 1 and type 2 diabetic patients suggesting altered axonal sodium (Na+) conductance and axonal sodium–potassium (Na+/K+) pump dysfunction [18–21]. These abnormalities are present in patients with established diabetic neuropathy [12], correlate strongly with neuropathy-related quality of life [22] and are consistent with animal models of diabetic neuropathy [6,9,23]. Recent studies have demonstrated that these changes are also present prior to the development of clinical neuropathy [16,24].

Treatment of diabetic peripheral neuropathy consists of alleviation of neuropathic symptoms and optimization of glycaemic control [25]. The Diabetes Control and Complications Trial and Epidemiology of Diabetes Interventions and Complications study found that strict glycaemic control through intensive insulin therapy achieved by multiple daily insulin injections (MDII) or continuous subcutaneous insulin infusion (CSII) was associated with reduced prevalence and delayed progression of diabetic peripheral neuropathy in T1D [26]. The benefits of CSII over MDII include reduction in the number of severe hypoglycaemic events, lowering of glucose exposure and improved treatment satisfaction for diabetic patients [27–31]. However, the potential differences between the two regimens in terms of their effect on peripheral axonal function have not been systematically investigated.

The major aim of this study was to utilize nerve excitability techniques to investigate whether there were differences in axonal function of patients with T1D treated with either CSII or MDII therapy. The secondary aim was to establish whether these changes could be detected prior to the onset of clinical symptoms or nerve conduction evidence of neuropathy. Lastly, we wanted to investigate if these abnormalities were present at longitudinal follow-up in a subgroup of tested patients.

Materials and methods

A total of 55 patients with T1D were consecutively recruited and assessed at the Prince of Wales Hospital in Sydney. The study was approved by the South Eastern Sydney Area Health Service and the University of New South Wales, and all patients gave written informed consent in agreement with the Declaration of Helsinki. Diagnosis of DM was made by endocrinologists on the basis of clinical characteristics, age, weight and family history, and where further confirmation was required, presence of anti-GAD antibodies and c-peptide tests were conducted. All patients had been on their respective insulin treatment type for a minimum of 12 months prior to testing. Clinical characteristics were obtained, including body mass index, duration of diabetes diagnosis, random blood glucose by needle stick at the time of testing (Accu-Check Performa®, Roche Diagnostics, Indianapolis, USA) and HbA1c % at the time of testing and for 2 years prior to assessment. All patients underwent a comprehensive neurological examination to determine the presence of diabetic neuropathy by an examiner blinded to treatment type. This included assessment of sensation to pin prick, vibration, strength tests, symptom report and standard nerve conduction studies: sural amplitude, sural velocity, tibial amplitude and tibial latency assessment (Medelec Synergy system, Oxford Instruments, UK). The presence of retinopathy and nephropathy was also assessed at or within 1 month of testing per standard care at the hospital.

Patients without clinical signs of neuropathy (n = 41) then underwent assessment of axonal ion channel function using nerve excitability techniques. Excitability testing was performed at initial appointments and at 6-12 months follow-up in a subgroup of patients (n = 18). Patient clinical and nerve excitability assessment results were then grouped and compared according to insulin regimen: CSII (n = 17) and MDII (n = 24). MDII treatment consisted of at least three injections per day of rapid acting 9 Aspart, Lispro and either glargine and detemir. CSII treatment involved a basal infusion of insulin with a large bolus infusion at meals. Twenty age-matched and gender-matched normal controls were also recruited and underwent nerve excitability assessments for comparison with patient results.

Assessment of axonal ion channel function

All subjects underwent nerve excitability testing, which was undertaken by an investigator blinded to treatment allocation. Skin temperature was monitored close to the site of testing. Testing was performed by stimulating the median nerve at the wrist and measuring the compound muscle action potential (CMAP) of abductor pollicis brevis using surface electrodes (Unomedical, Bikerod, Denmark). QTRAC automated software (Digitimer, London, UK) was used to apply the TRONDNF protocol [32]. The software allowed rapid acquisition of nerve excitability properties pertaining to five distinct testing paradigms: stimulus response (SR) behaviour, strength–duration time constants (SDTC), threshold electrotonus (TE), current threshold relationships (I/V) and the recovery cycle (RC).
To obtain SR curves, investigators applied a 1-ms duration current of increasing intensity until the maximal CMAP response was established. In the remaining tests, a target of ~40% maximum CMAP was utilized, and the stimulus required to achieve this target was termed ‘threshold’. SDTC was established with Weiss’ Law [33] as the relationship between strength and duration of a stimulus using four stimulus durations (0.2, 0.4, 0.8 and 1 ms). This time constant is reflective of nodal persistent Na+ conductance [34]. TE was determined by plotting the percentage of threshold change when 1-ms test pulses were applied during and after 100-ms sub-threshold conditioning currents of +40% (depolarizing—TEd) and −40% (hyperpolarizing—TEh) control threshold. TE provides information on internodal properties and overall axonal membrane potential [17]. I/V was determined by mapping change in threshold when 1-ms test impulses were delivered following 200-ms depolarizing and hyperpolarizing conditioning currents (+50 to −100 ms). I/V provides information regarding rectification properties of the internode [35,36]. The RC assesses the change in threshold that occurs over 200 ms following supramaximal stimulation. It provides information on Na+ and K+ channels at the nodal and paranodal regions of the axon [35].

Statistical analysis

Statistical analysis was performed using spss statistics software v. 20 (IBM, Chicago, IL, USA). Assessments of normality were first undertaken on patient group data using the Shapiro Wilk test. To determine if axonal dysfunction occurs in non-neuropathic patients with T1D, nerve excitability results from all non-neuropathic patients (n = 34) were compared with those from controls (n = 20) using student t-tests or Mann–Whitney U-tests where appropriate. To establish if there were differences in axonal function between patients treated with MDII and those treated with CSII, nerve excitability parameters from the MDII-treated and CSII-treated groups were compared with each other and normal controls. To investigate potential relationships between clinical characteristics of interest, nerve conduction studies and neurophysiological measures, Spearman rank or Pearson’s correlations were undertaken where appropriate. Mean values where provided are expressed as mean ± SE. Findings were considered statistically significant when P < 0.05.

Results

The clinical and demographic characteristics for the patients (n = 41) are shown in Table 1. Seventeen patients were receiving CSII therapy and 24 were receiving MDII. Patients receiving CSII had all previously been treated with a standard MDII regimen at the time of initial diagnosis and subsequently switched to CSII therapy. The average duration of CSII treatment was 5.38 ± 1.43 years. No correlations were found between neurophysiology parameters and duration of CSII therapy. There were also no significant differences in glycaemic control, duration of diabetes, age, gender, body mass index, total insulin requirement or sensory or motor amplitudes, latencies or velocities between the two treatment groups Sural nerve velocities were trending towards faster in the CSII group. Of note, five patients receiving MDII treatment presented with ophthalmic evidence of mild but stable retinopathy compared with only one patient treated with CSII. No signs of nephropathy were noted in the patient cohort as established by an estimated glomerular filtration rate >90 mL/kg/m² and no evidence of microalbuminuria. Clinically notable hypoglycaemic events were reported by three patients (two CSII and one MDII) within the week prior to assessment.

When compared with age-matched and gender-matched normal controls (n = 20), the patients as a group demonstrated altered axonal nerve excitability parameters (Table 2). Of note, there was a reduced percentage change in TEh at 90–100 ms (patients −110.40 ± 2.41, controls −121.20 ± 3.89, P < 0.05) and TEd undershoot

Table 1. Clinical characteristics for type 1 diabetic patients as group and according to insulin treatment regimen

<table>
<thead>
<tr>
<th>Treatment type</th>
<th>All patients n = 41</th>
<th>MDII n = 24</th>
<th>CSII n = 17</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>M : F</td>
<td>23:18</td>
<td>14:10</td>
<td>9:8</td>
<td>0.33</td>
</tr>
<tr>
<td>Age (years)</td>
<td>27.66 ± 1.37</td>
<td>27.50 ± 0.40</td>
<td>27.88 ± 2.71</td>
<td>0.33</td>
</tr>
<tr>
<td>Disease duration (months)</td>
<td>119.69 ± 15.68</td>
<td>112.86 ± 19.32</td>
<td>130.43 ± 25.25</td>
<td>0.57</td>
</tr>
<tr>
<td>HbA1c %</td>
<td>7.76 ± 0.21</td>
<td>7.74 ± 0.27</td>
<td>7.79 ± 0.34</td>
<td>0.92</td>
</tr>
<tr>
<td>(2 year average)</td>
<td>(8.30 ± 0.28)</td>
<td>(8.51 ± 0.35)</td>
<td>(7.92 ± 0.46)</td>
<td>0.21</td>
</tr>
<tr>
<td>RBG (mmol/L)</td>
<td>10.13 ± 0.97</td>
<td>10.64 ± 1.40</td>
<td>9.35 ± 1.22</td>
<td>0.48</td>
</tr>
<tr>
<td>Body mass index (kg/m²)</td>
<td>25.29 ± 0.69</td>
<td>25.93 ± 0.79</td>
<td>26.09 ± 1.28</td>
<td>0.73</td>
</tr>
<tr>
<td>Average daily insulin (units)</td>
<td>51.97 ± 0.97</td>
<td>54.45 ± 28.45</td>
<td>48.32 ± 15.63</td>
<td>0.74</td>
</tr>
<tr>
<td>Sural amplitude (µV)</td>
<td>18.44 ± 0.97</td>
<td>19.60 ± 1.37</td>
<td>17.11 ± 0.32</td>
<td>0.54</td>
</tr>
<tr>
<td>Sural conduction velocity (m/s)</td>
<td>43.72 ± 7.30</td>
<td>41.51 ± 1.45</td>
<td>46.75 ± 1.48</td>
<td>0.07</td>
</tr>
<tr>
<td>Tibial amplitude (mV)</td>
<td>15.68 ± 0.97</td>
<td>15.32 ± 1.23</td>
<td>16.16 ± 1.10</td>
<td>0.44</td>
</tr>
<tr>
<td>Tibial latency (ms)*</td>
<td>3.35 ± 0.06</td>
<td>3.49 ± 0.05</td>
<td>3.16 ± 0.07</td>
<td>0.19</td>
</tr>
</tbody>
</table>

Values are given as mean ± SEM. CSII, continuous subcutaneous insulin infusion; MDII, multiple daily insulin injections; DOD, duration of diabetes; RBG, random blood glucose. CSII versus MDII: *P < 0.05.
Table 2. Nerve excitability values for the treatment groups and controls at initial assessment and 6–12 months follow-up

<table>
<thead>
<tr>
<th>Excitability parameters</th>
<th>All patients</th>
<th>MDII</th>
<th>CSII</th>
<th>Controls</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n = 41</td>
<td>n = 24</td>
<td>n = 17</td>
<td>n = 20</td>
</tr>
<tr>
<td>Initial assessment</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Threshold electrotonus</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TEd (10–20 ms)</td>
<td>65.22 ± 0.70*</td>
<td>63.16 ± 0.78**</td>
<td>68.13 ± 3.89</td>
<td>67.80 ± 1.08</td>
</tr>
<tr>
<td>TEd (accommodating)</td>
<td>20.73 ± 0.53*</td>
<td>19.58 ± 0.53**</td>
<td>22.35 ± 0.92</td>
<td>22.81 ± 0.83</td>
</tr>
<tr>
<td>TEh (undershoot)</td>
<td>−16.46 ± 0.45**</td>
<td>−15.74 ± 0.53**</td>
<td>−17.47 ± 0.71</td>
<td>−18.98 ± 0.74</td>
</tr>
<tr>
<td>TEh (90–100 ms)</td>
<td>−110.40 ± 2.41*</td>
<td>−103.80 ± 2.47**</td>
<td>−119.60 ± 3.67</td>
<td>−121.20 ± 3.89</td>
</tr>
<tr>
<td>Recovery cycle</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Superexcitability</td>
<td>−22.18 ± 0.01*</td>
<td>−22.17 ± 0.84**</td>
<td>−24.12 ± 0.08</td>
<td>−26.47 ± 1.20</td>
</tr>
<tr>
<td>Subexcitability</td>
<td>11.15 ± 0.53**</td>
<td>10.14 ± 0.44**</td>
<td>12.51 ± 1.03</td>
<td>15.02 ± 0.03</td>
</tr>
<tr>
<td>Follow up</td>
<td>n = 18</td>
<td>n = 12</td>
<td>n = 6</td>
<td>n = 20</td>
</tr>
<tr>
<td>Threshold electrotonus</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TEd (10–20 ms)</td>
<td>64.76 ± 1.09</td>
<td>63.21 ± 0.85**</td>
<td>68.70 ± 2.11</td>
<td>67.80 ± 1.08</td>
</tr>
<tr>
<td>TEd (undershoot)</td>
<td>−16.01 ± 0.70*</td>
<td>−16.27 ± 0.64*</td>
<td>−18.98 ± 0.74</td>
<td>−18.98 ± 0.74</td>
</tr>
<tr>
<td>TEh (90–100 ms)</td>
<td>−111.00 ± 0.01</td>
<td>−102.06 ± 0.64*</td>
<td>−121.02 ± 6.62</td>
<td>−121.20 ± 3.89</td>
</tr>
<tr>
<td>Recovery cycle</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Superexcitability</td>
<td>−23.82 ± 1.02*</td>
<td>−22.98 ± 1.35**</td>
<td>−26.47 ± 1.20</td>
<td>−26.47 ± 1.20</td>
</tr>
<tr>
<td>Subexcitability</td>
<td>−10.54 ± 0.74*</td>
<td>9.76 ± 0.79*</td>
<td>12.00 ± 1.45</td>
<td>15.02 ± 0.03</td>
</tr>
</tbody>
</table>

Values are expressed as percentage change in threshold (mean ± SEM). All axonal excitability parameters were normally distributed. The MDII-treated group show significant abnormality across numerous parameters when compared with CSII-treated group and controls.

These changes are indicative of axonal membrane depolarization, CSII, continuous subcutaneous insulin infusion; MDII, multiple daily insulin injections; TEd, depolarizing threshold electrotonus; TEh, hyperpolarizing threshold electrotonus.

Changes were also noted in recovery cycle parameters, specifically superexcitability (%) which was reduced in patients (−22.18 ± 0.01) compared with controls (−26.47 ± 1.20, P < 0.05) and reduced subexcitability (patients 11.15 ± 0.53, controls 15.02 ± 0.03, P < 0.01). The pattern of change in excitability parameters is consistent with axonal membrane depolarization and agrees with previous studies in patients with T1D.

When analysed according to treatment type, prominent abnormalities of axonal function were noted in the MDII group compared with both CSII-treated patients and normal controls (Table 2). Specifically, in the MDII group, reduced threshold change (%) and concurrent flattening of mean plots (Figure 1A and B) was noted in the recovery cycle parameters superexcitability (MDII −22.17 ± 0.84, controls −26.47 ± 1.20, P < 0.01) and subexcitability (MDII 10.14 ± 0.44, controls 15.02 ± 0.03, P < 0.01). Likewise, reduced threshold change in TEd and TEh was found at multiple time points: TEd (10–20 ms) (MDII 63.16 ± 0.78, controls 67.80 ± 1.08, P < 0.01), TEd undershoot (MDII −15.74 ± 0.53, controls −18.98 ± 0.74, P < 0.01) and TEh (90–100 ms) (MDII −103.80 ± 2.47, controls −121.20 ± 3.89, P < 0.01) (Figure 1B). Additionally, compared with the CSII-treated group, MDII-treated patients had significantly reduced threshold change in TEd (10–20 ms) (CSII 68.13 ± 3.89, P < 0.05), TEd (accommodating) (CSII 22.35 ± 0.92, P < 0.05), TEh (90–100 ms) (CSII −119.60 ± 3.67, P < 0.01), superexcitability (CSII −24.12 ± 1.08, P < 0.05) and subexcitability (CSII 12.51 ± 1.03, P < 0.05) (Table 2).

In contrast to the MDII-treated group, the CSII-treated group demonstrated no significant change in excitability parameters compared with controls (Table 2). Specifically, there were no significant changes in TE parameters or in measures of the recovery cycle, with mean data plots for CSII-treated patients showing relative normality compared with control plots (Figures 1C and D).

Longitudinal assessment of axonal function

Longitudinal studies were undertaken at 6–12 months on a subgroup of the patient cohort (n = 18). Over this period, there had been no significant change in clinical features or measures of glycaemic control in either group. The previously demonstrated abnormalities in excitability parameters persisted in the MDII group (n = 12) (Table 2). Significant abnormalities were again noted in TE and recovery cycle parameters. In TE, TEd (10–20 ms), TEd (undershoot) and TEh (90–100 ms) (P < 0.05) (Figure 2A and B) were significantly different in MDII-treated patients compared with controls with TEd (10–20 ms) also showing reductions compared with the CSII subgroup at follow-up (CSII 68.70 ± 2.11, P < 0.05). Likewise, recovery cycle parameters superexcitability (MDII −22.98 ± 1.35,
Discussion

The present study has demonstrated that patients with T1D that are treated with MDII have abnormal axonal function when compared with age-matched controls and CSII-treated patients. These changes were noted in cross-sectional studies and remained at longitudinal follow-up. In addition, faster lower limb sensory and motor nerve conduction was noted in CSII-treated patients, demonstrating that CSII is associated with improved nerve function in those regions that are most susceptible to the development of neuropathy, namely the distal lower limbs. All patients had been receiving their respective treatment type for at least 12 months prior to enrolment, and the choice of treatment was based largely on patient preference or, in some cases, advised by the treating physician because of the occurrence of nocturnal hypoglycaemic episodes. However, although patients were assessed by an investigator blinded to treatment, the lack of random assignment to treatment remains a potential limitation of the study.

The present study demonstrated that MDII-treated patients had significant abnormalities of excitability in contrast to CSII-treated patients (Table 2). Prominent changes were noted across a number of TE and recovery cycle parameters in a pattern that is consistent with depolarization of the axonal membrane potential [36]. The potential basis for membrane depolarization may relate to dysfunction of the energy-dependent axonal Na+/K+ pump.
pump. The Na\(^+\)/K\(^+\) pump is responsible for maintaining a normal axonal electrochemical gradient, and reduced activity of the Na\(^+\)/K\(^+\) pump has been demonstrated in nerve preparations from type 1 and type 2 diabetic patients [11]. Studies in animal models of T1D have also suggested that Na\(^+\)/K\(^+\) pump dysfunction may occur as a result of upregulated polyol pathway activity in the presence of hyperglycaemia, oxidative stress and c-peptide deficiency [14,37,38].

As CSII is associated with improved glycaemic control [30,39], it could be argued that the differences in axonal function were related to better glycaemic control or differences in the total amounts of insulin administered in the two groups. However, the present study failed to establish differences in glycaemic control between the two treatment groups or to find correlations between HbA\(_1c\) % and nerve excitability parameters. Previous work has provided evidence of changes in nerve conduction velocity and intraepidermal nerve fibre with alterations in insulin dosing, independent of changes in glucose levels [25,40,41]. However, there were no significant differences between the two treatment groups in terms of total insulin requirement in the present study. Additionally, no correlations were found between duration of insulin treatment through CSII and neurophysiological parameters.

An alternative explanation for the differences between treatment groups may relate to differences in blood glucose variability, which has been linked to overall morbidity [42], microvascular dysfunction [43] and oxidative stress in diabetic subjects [44–46]. It is well established that oxidative stress is well linked to complications in diabetes [47] in particular endothelial dysfunction [48] and subsequently compromised microvasculature function [49]. Previous studies have found less glucose variability in patients treated with CSII compared with MDII [50]. We speculate that the CSII-treated patients in the present study may have had a reduced variability in glucose profile than the MDII-treated patients, thus protecting CSII-treated patients from the consequences of microvascular dysfunction and oxidative stress. The present study did not quantify glucose variability; however, further studies incorporating continuous glucose monitoring would be needed to assess the effects of glucose variability and direct insulin signalling on axonal function.

From a clinical perspective, the findings of the present investigation have important implications. Although the duration of follow-up in the present study was limited to 12 months in a subgroup of the initial cohort, longer periods of assessment in a larger cohort may help delineate whether the better axonal function in CSII-treated patients results in better clinical outcomes. The basis for this relates to the previously defined association between nerve ion channel dysfunction and axonal degeneration [51]. Chronic changes in axonal ion channel function have been shown to trigger a cascade of events resulting in reverse operation of Na\(^+\)/Ca\(^+\) exchanger, causing an excitotoxic Ca\(^+\) influx and subsequent axonal degeneration [51,52]. This study also suggests that nerve excitability techniques may be able to identify a window of opportunity for which therapeutic interventions targeting axonal ion channel function may be useful. Although the present study established differences and magnitude of these differences in axonal function between CSII and MDII, further investigation to clarify the effects of insulin treatment regimens is required using larger randomized control trials and crossover studies.

In total, the findings of the present study suggest that non-neuropathic type 1 diabetic patients treated with CSII have normal axonal ion channel function compared with MDII-treated patients with similar clinical characteristics. The altered axonal function noted in MDII-treated patients was similar to that seen in patients with established diabetic neuropathy and is indicative of axonal membrane depolarization. The findings of this study are relevant clinically as they raise the possibility that CSII therapy may have neuroprotective potential in T1D. These data provide the basis for further investigation of the differential effects of insulin treatment on axonal function in a clinical trial.

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Author contributions

N.K. was involved in study design, recruitment, data collection, data interpretation and manuscript composition. R.A. contributed to recruitment data collection, discussion and manuscript composition. A.P. was involved in recruitment, discussion and manuscript composition. C.L. was involved in interpretation of data. M.K. was involved in interpretation of data and manuscript drafting. A.K. was involved in study design, data interpretation and manuscript composition.

Conflicts of interest

The authors have no conflicts of interest.

Guarantor’s statement

Associate Professor Arun V. Krishnan is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.
Neuroprotective Potential of CSII

References

ess Centres, ACT, 2010; 174.
4. Tavakoli M, Kallinikos PA, Efron N, Boulton AJM, Malik RA. Corneal sen- sitivity is reduced and relates to the severity of neuropathy in patients with 
7. Bierhaus A, Fleming T, Stoyanov S, Black JA, Waxman SG. Changes of sodium channel expression in experimen- 
8. Greene DA, Yagihashi S, Lattimer SA. Axonal potassium conductance and 
9. Bhat NA, Kallinikos PA, Efron N, Boulton AJM, Malik RA. Corneal sen- 
sitivity is reduced and relates to the severity of neuropathy in patients with 
avon analogs versus insulin pump therapy for the treatment of type 1 and type 2 diabe- 
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