

RESEARCH

Open Access

# Corneal confocal microscopy differentiates inflammatory from diabetic neuropathy



Michael Fleischer<sup>1</sup>, Inn Lee<sup>2</sup>, Friedrich Erdlenbruch<sup>1</sup>, Lena Hinrichs<sup>3</sup>, Ioannis N. Petropoulos<sup>4,5</sup>, Rayaz A. Malik<sup>4,5</sup>, Hans-Peter Hartung<sup>2,6,7,8</sup>, Bernd C. Kieseier<sup>2</sup>, Christoph Kleinschnitz<sup>1</sup> and Mark Stettner<sup>1\*</sup>

## Abstract

**Background:** Immune-mediated neuropathies, such as chronic inflammatory demyelinating polyneuropathy (CIDP) are treatable neuropathies. Among individuals with diabetic neuropathy, it remains a challenge to identify those individuals who develop CIDP. Corneal confocal microscopy (CCM) has been shown to detect corneal nerve fiber loss and cellular infiltrates in the sub-basal layer of the cornea. The objective of the study was to determine whether CCM can distinguish diabetic neuropathy from CIDP and whether CCM can detect CIDP in persons with coexisting diabetes.

**Methods:** In this multicenter, case-control study, participants with CIDP ( $n = 55$ ) with ( $n = 10$ ) and without ( $n = 45$ ) diabetes; participants with diabetes ( $n = 58$ ) with ( $n = 28$ ) and without ( $n = 30$ ) diabetic neuropathy, and healthy controls ( $n = 58$ ) underwent CCM. Corneal nerve fiber density (CNFD), corneal nerve fiber length (CNFL), corneal nerve branch density (CNBD), and dendritic and non-dendritic cell density, with or without nerve fiber contact were quantified.

**Results:** Dendritic cell density in proximity to corneal nerve fibers was significantly higher in participants with CIDP with and without diabetes compared to participants with diabetic neuropathy and controls. CNFD, CNFL, and CNBD were equally reduced in participants with CIDP, diabetic neuropathy, and CIDP with diabetes.

**Conclusions:** An increase in dendritic cell density identifies persons with CIDP. CCM may, therefore, be useful to differentiate inflammatory from non-inflammatory diabetic neuropathy.

**Keywords:** Chronic inflammatory demyelinating neuropathy, corneal confocal microscopy, Diabetes mellitus

## Background

Immune-mediated neuropathies are a heterogeneous group of conditions mediated by inflammatory processes resulting in impaired sensation and muscle weakness which impose a significant burden of disease [1]. The most common immune-mediated neuropathy is chronic inflammatory demyelinating polyneuropathy (CIDP), with a prevalence of around 1–2/100,000 people [2]. The diagnosis of CIDP relies primarily on nerve conduction

studies; however, diagnostic challenges can lead to a considerable degree of mis- and under-diagnosis [3, 4]. In some cases, neurophysiology may not clearly differentiate CIDP from other demyelinating neuropathies, such as demyelinating diabetic neuropathy or demyelinating hereditary neuropathies [5]. Furthermore, epidemiological data suggest an increased prevalence of CIDP among individuals with diabetes [6]. This, coupled with the difficulty of diagnosing CIDP in persons with diabetic neuropathy, may preclude the application of timely and specific treatment options such as intravenous immunoglobulins for CIDP [7].

\* Correspondence: [mark.stettner@uk-essen.de](mailto:mark.stettner@uk-essen.de)

<sup>1</sup>Department of Neurology and Center for Translational and Behavioral Neurosciences (C-TNBS), University Hospital Essen, University Duisburg-Essen, Essen, Germany

Full list of author information is available at the end of the article



© The Author(s). 2021 **Open Access** This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <http://creativecommons.org/licenses/by/4.0/>. The Creative Commons Public Domain Dedication waiver (<http://creativecommons.org/publicdomain/zero/1.0/>) applies to the data made available in this article, unless otherwise stated in a credit line to the data.

Corneal confocal microscopy (CCM) is a relatively rapid non-invasive ophthalmic imaging technique that is reproducible [8, 9] and well tolerated [10]. This technique has demonstrated nerve fiber loss and provided good diagnostic utility for diabetic neuropathy in larger cohorts [11–13], comparable to intraepidermal nerve fiber density [14] and also has shown predictive utility for the development of diabetic neuropathy [15]. CCM also identifies corneal nerve loss in a range of peripheral neuropathies including idiopathic small fiber neuropathy [16], Charcot-Marie-Tooth disease type 1A [17], HIV neuropathy [18], chemotherapy-induced peripheral neuropathy [19], amyloid neuropathy [20], and Friedreich's ataxia [21].

There is currently no non-invasive technique that can act as a surrogate measure of ongoing inflammation to assess disease progression or treatment response and dose adjustment of therapies. Experimental data indicate that the presence of dendritic cells and their contact with the sub-basal nerve plexus may trigger nerve fiber damage [22]. More recently, we have shown an increase in corneal dendritic cells in individuals with CIDP and suggested that this may help to stratify CIDP subtypes, clinical course, and disease activity [23]. In a prospective study of 17 individuals with CIDP who were followed over 18 months, the presence of > 30 cells/mm<sup>2</sup> at baseline identified clinical progression with a sensitivity and specificity of 100% [24]. Moreover, in one person with anti-neurofascin-155 neuropathy, treatment with rituximab was associated with a reduction in the serum antibody titer with a clinical and electrophysiological improvement and reduction of corneal inflammatory cell infiltrates [25].

The present study was undertaken to assess whether CCM can differentiate CIDP from diabetic neuropathy and whether it can detect CIDP in persons with coexisting diabetes.

## Methods

### Study design and participants

The study was performed in accordance with the principles of the Declaration of Helsinki, and the local Ethics Committees approved the study plan (Ethics Committee University of Essen, #16-7289-BO, North Manchester Ethics Committee). Participants who provided written informed consent were included. All participants were over 18 years of age. A total of 55 patients with CIDP, 58 patients with type 2 diabetes and 58 healthy controls were investigated. Within the CIDP group, 10 patients also had type 2 diabetes, and 11 had monoclonal gammopathy of undetermined significance (MGUS+), (Table 1). CIDP was diagnosed according to the European Federation of Neurological Societies/Peripheral Nerve Society criteria [26]. Patients with diabetes underwent assessment of the

**Table 1** Subgroups and demographics of participants.

| Cohort (total <i>n</i> = 171) | <i>n</i> | Age       | Sex Male (%) | Female (%) |
|-------------------------------|----------|-----------|--------------|------------|
| CIDP                          | 55       | 58 ± 12.7 | 60           | 40         |
| CIDP + diabetes               | 10       | 55 ± 8.3  | 90           | 10         |
| CIDP –diabetes                | 45       | 59 ± 12.3 | 53           | 47         |
| CIDP + MGUS                   | 11       | 58 ± 9.3  | 66           | 34         |
| Diabetes                      | 58       | 52 ± 16.7 | 47           | 53         |
| Diabetes +to                  | 28       | 61 ± 10.8 | 43           | 57         |
| Diabetes –to                  | 30       | 43 ± 16.6 | 47           | 53         |
| Healthy controls              | 58       | 49 ± 15.3 | 34           | 66         |

*Abbreviations:* CIDP chronic inflammatory demyelinating polyneuropathy, MGUS monoclonal gammopathy of undetermined significance, DN diabetic neuropathy, patients with (+to) and without (–to) neuropathy according to the Toronto criteria

neuropathy symptom profile (NSP) and modified neuropathy disability score (NDS) to assess pinprick, vibration perception, temperature sensation and ankle reflexes. Nerve conduction studies (NCS) were undertaken and diabetic neuropathy was defined according to the Toronto consensus, which requires the presence of symptoms (abnormal NSP) or signs of neuropathy (NDS > 2) and abnormal peroneal nerve conduction velocity (PMNCV < 40 m/s) and the patients were subdivided into those with diabetic neuropathy (*n* = 28) and those without (*n* = 30) [27].

Patients with CIDP who were positive for anti-MAG antibodies were excluded. In the healthy control group, a full blood workup and clinical, neurological, and neurophysiological examination were performed to exclude neuropathy. Patients and controls were recruited from the Department of Neurology, University Hospital of Essen, Germany, and from the Centre for Endocrinology and Diabetes, University of Manchester, UK.

### Corneal confocal microscopy

Corneal images were captured using a Heidelberg Retina Tomograph (HRT III, Rostock Cornea Module, Heidelberg Engineering, Heidelberg, Germany). Corneal integrity was confirmed by slit-lamp examination. Local anesthetic (0.4% benoxinate hydrochloride) was used to anesthetize the eye, and a drop of Viscotears Liquid Gel was used between the lens and the disposable lens cover. CCM is a corneal contact technique which has a very low risk for corneal injury or keratitis; however, none of our patients developed any of these complications. Four scan cycles were conducted across the entire depth of the central cornea, especially the sub-basal nerve layer. At least 15 images per patient, meeting established quality criteria were analyzed [10]. Automated corneal nerve quantification was undertaken using established software (ACCMetrics Image Analysis tool v1.1, University of Manchester, UK) to evaluate the following: corneal nerve

fiber density (CNFD; no./mm<sup>2</sup>), corneal nerve fiber length (CNFL; mm/mm<sup>2</sup>), and corneal nerve branch density (CNBD; major no./mm<sup>2</sup>). Cell quantification was performed in a blinded manner without knowledge of patient diagnosis using ImageJ software (version 1.41, National Institutes of Health, USA). Cells were classified as dendritic cells with fiber contact (DCF), dendritic cells in the periphery without fiber contact (DCP), non-dendritic cells with fiber contact (NCF), or non-dendritic cells in the periphery without fiber contact (NCP), as described previously [23]. Dendritic and non-dendritic cells were counted per mm<sup>2</sup>. F/mm<sup>2</sup> comprises all cells/mm<sup>2</sup> with fiber contact (DCF and NCF), whereas P/mm<sup>2</sup> combines all cells per mm<sup>2</sup> without fiber contact (DCP, NCP).

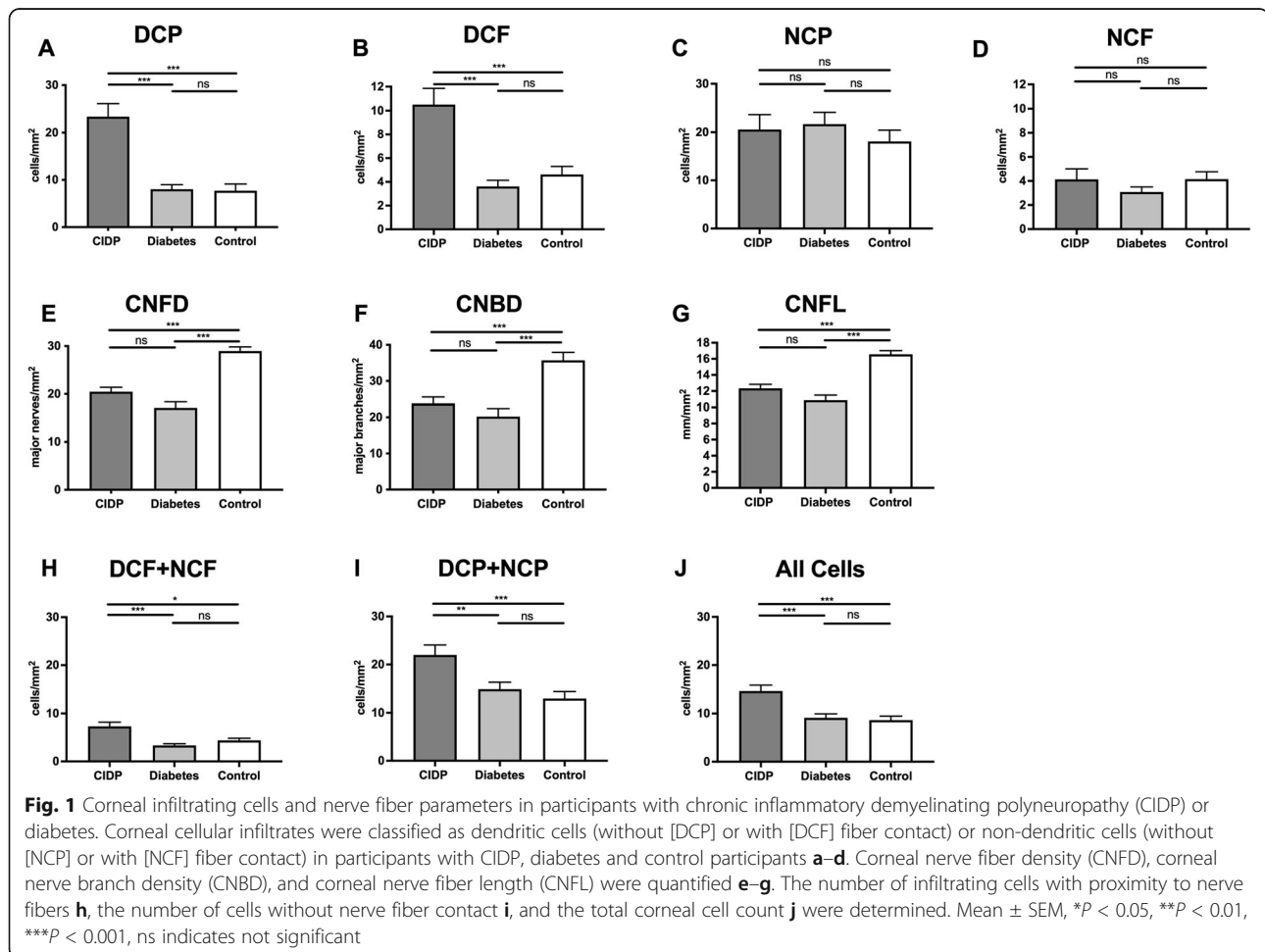
### Statistical analysis

All data are presented as mean, standard error of the mean, and *P* values, which were calculated using GraphPad Prism software version 9.0 (GraphPad Software, Inc., La Jolla, CA, USA). Differences between groups were assessed using Kruskal-Wallis one-way analysis of

variance with Dunn's multiple comparison post hoc tests, after analyzing for parametrical distribution with Shapiro-Wilk test. A *P* value < 0.05 was considered to be significant (\* < 0.05, \*\* < 0.01, and \*\*\* < 0.001). Specificity, sensitivity, and positive predictive value were calculated for distinguishing CIDP from DN and healthy controls with the parameter DCP and DCF by using the lower *z* value of two times the SEM from the median as the cut-off value.

### Results

The mean age of patients with CIDP was 58 ± 12.7 years and 60% were males, compared to the group of patients with diabetes who had a mean age of 52 ± 16.7 years and 47% were males. The groups and subgroups (CIDP ± diabetes, CIDP ± MGUS, diabetes ± to) were matched with regard to demographic characteristics and did not differ significantly. The mean age of healthy controls was 49 ± 15.3 years and 58% were males (Table 1). The potential effect of age or sex as a confounding factor was examined by multiple regression analysis (method enter)



and logistic regression analysis. Neither factor was shown to have any influence on relevant parameters.

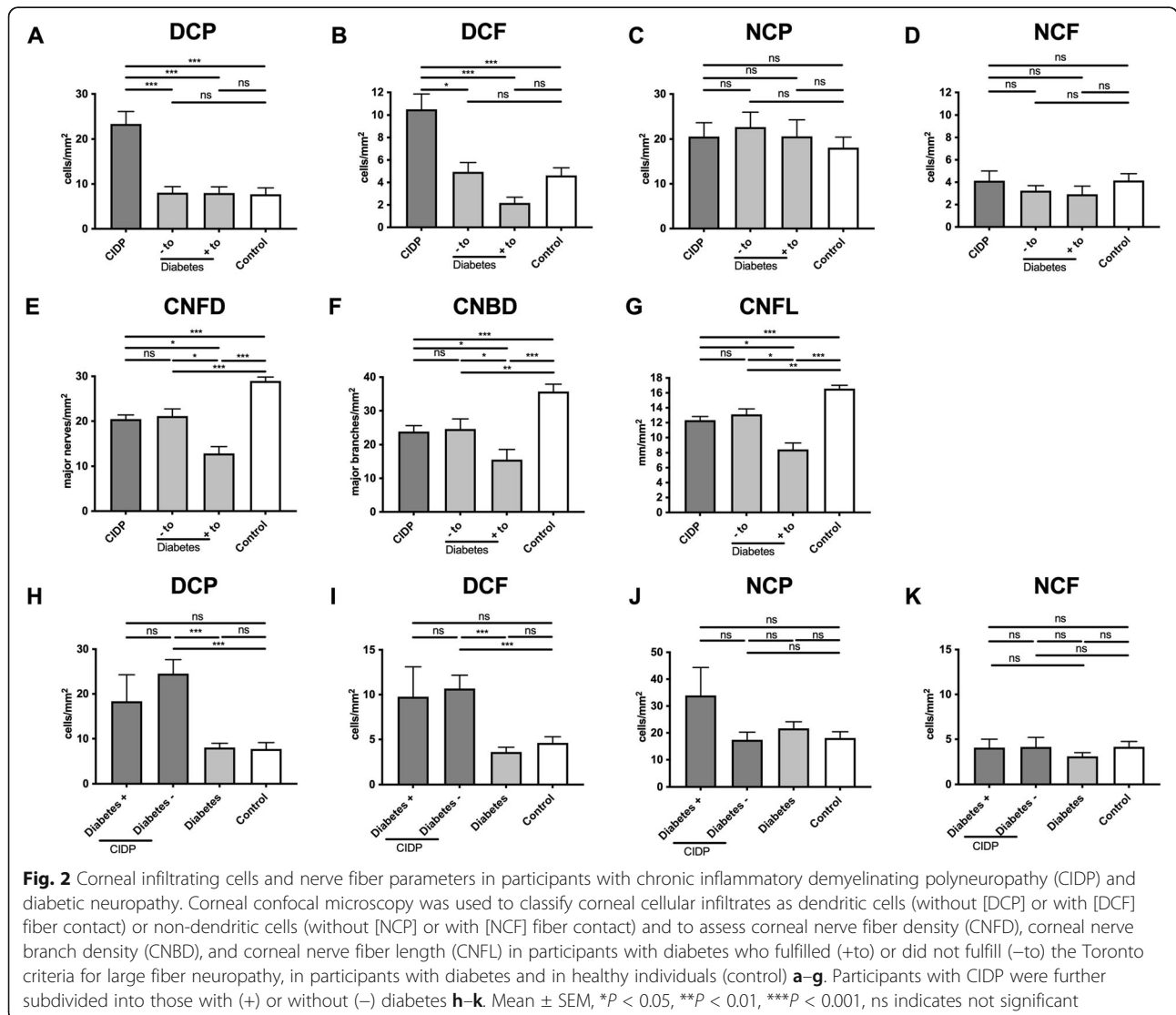
**CIDP and diabetes**

Patients with CIDP had a significantly higher DCP and DCF compared to patients with diabetes and healthy controls (Fig. 1a, b). There were no significant differences among groups for NCP (Fig. 1c) or NCF (Fig. 1d). However, all corneal nerve fiber parameters (CNFD, Fig. 1e; CNBD, Fig. 1f; CNFL, Fig. 1g) were significantly reduced in patients with CIDP and diabetes compared to controls, with no significant difference between CIDP and diabetes. The number of infiltrating cells in proximity to nerve fibers (DCF + NCF, Fig. 1h), the number of cells without nerve fiber contact (DCP + NCP, Fig. 1i), and the total cell number (Fig. 1j) was significantly higher in patients

with CIDP compared to patients with diabetes or compared to control.

To test whether the detection of CIDP could be further improved, as per pre-specified analysis, we normalized infiltrating cell numbers to nerve fiber parameters. For the ratio of total cell numbers to nerve fiber parameters, there was no significant improvement in identifying CIDP (Supplementary Figure 1a-c). There was a significant difference for cells in proximity to nerve fibers (DCF + NCF) and their ratio for CNFD, CNBD, and CNFL between patients with CIDP and diabetes (Supplementary Figure 1D-F). There was no significant difference for cells in the periphery (DCP + NCP) and their ratio to nerve fiber parameters between patients with CIDP and diabetes (Supplementary Figure 1g-i).

By comparing effect sizes (*d*), the ratio of DCF + NCF to CNFL was identified as the most selective



(F/mm<sup>2</sup>/CNFL  $d = 0.857$ ), followed by F/mm<sup>2</sup>/CNFD ( $d = 0.664$ ), F/mm<sup>2</sup>/CNBD ( $d = 0.408$ ), P/mm<sup>2</sup>/CNFL ( $d = 0.225$ ), P/mm<sup>2</sup>/CNFD ( $d = 0.083$ ), and P/mm<sup>2</sup>/CNBD ( $d = 0.079$ ). However, these derived parameters did not improve the ability to distinguish CIDP from DN beyond the number of infiltrating dendritic cells, which showed the highest effect size (DCF/mm<sup>2</sup>:  $d = 1.014$ ; DCP/mm<sup>2</sup>:  $d = 0.941$ ). DCP distinguished CIDP from DN with a sensitivity of 0.701, specificity of 0.879, and a PPV of 0.784. DCF distinguished CIDP from DN with a sensitivity of 0.596, specificity of 0.775, and a PPV of 0.723. For distinguishing CIDP from healthy control, DCP showed a sensitivity of 0.701, specificity of 0.767, and a PPV of 0.754, while DCF showed a sensitivity of 0.596, specificity of 0.722, and a PPV of 0.693.

**Patients with and without diabetic neuropathy**

Patients with diabetes were further subdivided into patients with (+to) and without (-to) neuropathy according to the Toronto criteria. Both +to and -to had significantly lower numbers of DCP and DCF compared to patients with CIDP (Fig. 2a, b). There were no significant differences between groups for NCP and NCF (Fig. 2c, d). CNFD, CNBD and CNFL were lower in +to compared to -to, and CNFD,

CNBD and CNFL were lower in -to group compared to controls (Fig. 2e-g).

**Influence of glycemia**

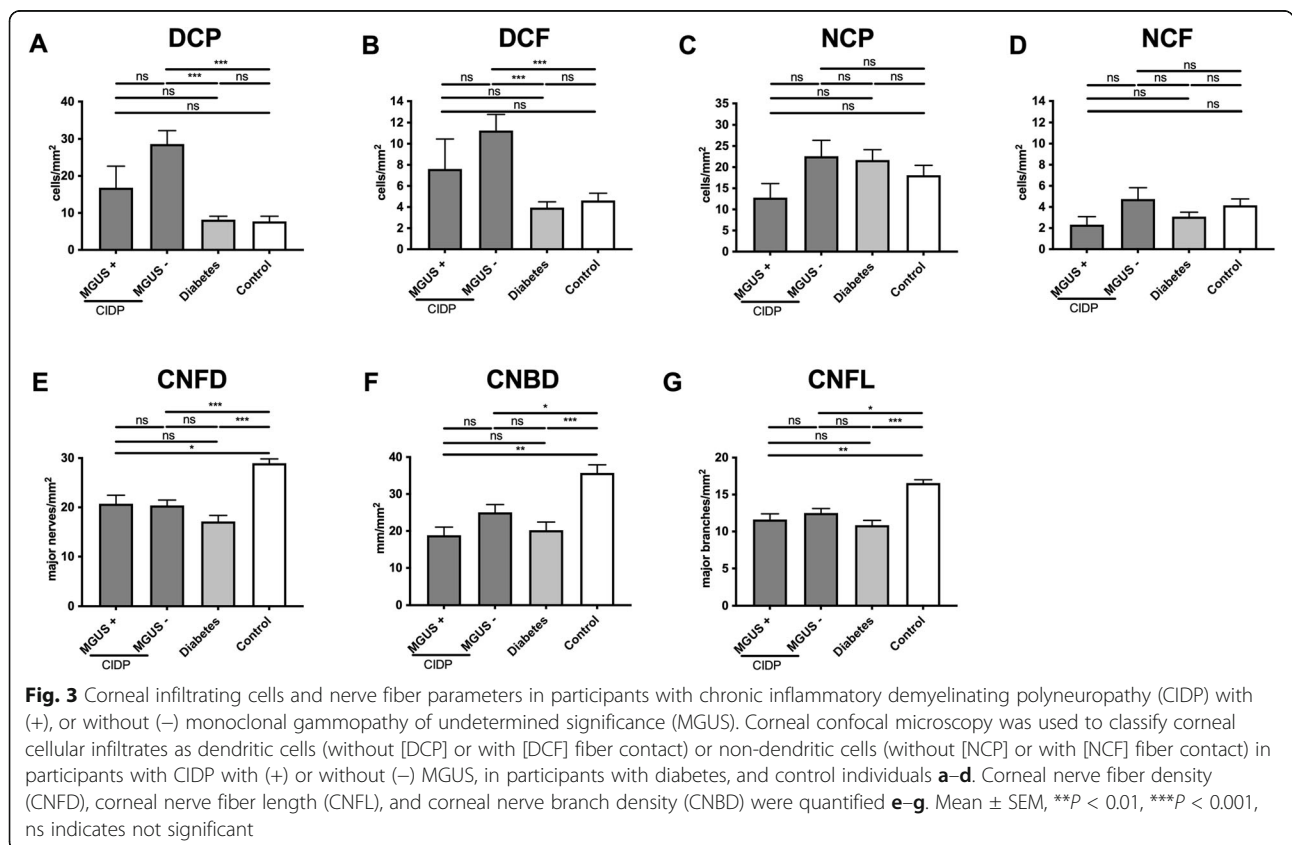
There was no significant difference in corneal cellular infiltrates in a comparison of participants with poor versus good glycemic control (HbA<sub>1c</sub> > 7.0% vs. ≤ 7.0%, Supplementary Figure 1j-k).

**CIDP and diabetes**

There were no significant differences in DCP, DCF, NCP, and NCF in a comparison of participants with CIDP with and without diabetes (Fig. 2h-k). DCP was significantly higher in the CIDP subgroup without diabetes but not in the CIDP subgroup with diabetes compared to participants with diabetes alone (Fig. 2h). CIDP individuals without diabetes had significantly higher numbers of DCF compared to individuals with diabetes alone (Fig. 2i). There were no significant differences for NCP and NCF (Fig. 2j, k) as well as for CNFD, CNFL, and CNBD in CIDP participants with or without diabetes (data not shown).

**MGUS neuropathy**

Participants with CIDP were divided into those with ( $n = 11$ ) and without ( $n = 44$ ) MGUS (Fig. 3).



Dendritic cell density (DCP and DCF) was significantly higher in the CIDP subgroup without MGUS compared to the diabetes or control groups (Fig. 3a, b). No significant differences were observed among any of the groups for NCP and NCF (Fig. 3c, d). CNFD, CNFL, and CNBD were significantly lower in the MGUS+, MGUS- and diabetes groups compared to controls and did not differ between MGUS+ and MGUS- groups (Fig. 3e–g).

## Discussion

Up until now, there has been no non-invasive method available to assess ongoing inflammation in the peripheral nervous system. Therefore, we aimed to investigate the potential of CCM to distinguish inflammatory from non-inflammatory neuropathies. We show an increase in corneal dendritic cells in proximity to corneal nerve fibers in participants with CIDP compared to diabetic neuropathy.

The prevalence of CIDP is increased among individuals with diabetes [7, 28]. However, the development of CIDP in persons with diabetes is often not diagnosed or misdiagnosed as diabetic neuropathy and these individuals may be denied timely and effective treatment [3]. None of the known biomarkers or diagnostic tools such as cerebrospinal fluid protein, clinical, electrophysiological, and serological markers of autoimmunity can adequately identify CIDP in persons with demyelinating neuropathy due to another condition such as diabetic neuropathy. Significant proximal motor involvement without autonomic involvement [29] and electrophysiological slowing may help to establish the diagnosis of CIDP as opposed to diabetic neuropathy [30–34]. Sural nerve biopsies can be used to identify inflammation but remain impractical as they are invasive and, therefore, of limited use in clinical practice [35, 36]. Immunohistochemical studies of cells in the cornea using cell-specific surface markers [37] indicate that the corneal dendritic cells are mainly Langerhans cells [38, 39] and a previous study has shown that contact between these dendritic cells and corneal nerves may trigger nerve fiber damage [22].

Our previous study showed corneal nerve loss and an increase in dendritic cells in proximity to corneal nerve fibers, which was associated with the severity of motor symptoms in CIDP [23].

Here, we show moderate diagnostic utility of CCM identification of increased corneal dendritic cell density to differentiate CIDP from diabetic neuropathy. This study also excluded HbA<sub>1c</sub> as a confounder in detecting CIDP among individuals with diabetes.

CCM was also useful to detect CIDP in those individuals with and without coexisting diabetes, indicating that

the increase in dendritic cells is driven by peripheral nerve inflammation. Indeed, recent longitudinal studies have shown that increased dendritic cell infiltration may predict worsening outcomes in CIDP [24] and infiltrates may be reduced in response to immune therapy [25].

Our data also show that dendritic cell density differs in CIDP subgroups with and without MGUS supporting the assertion that persons with CIDP and MGUS have different clinical and electrophysiological patterns and underlying pathophysiological mechanisms [40–42]. These findings are in keeping with our previous study where we also showed equivalent corneal nerve loss but lower numbers of corneal dendritic cells in those with MGUS compared to CIDP [23].

## Conclusion

Our study shows that corneal confocal microscopy, by enabling the quantification of corneal dendritic cells, may have clinical utility to differentiate inflammatory from diabetic neuropathy. However, larger longitudinal studies are required to evaluate the potential of this method to predict disease progression and response to treatment.

## Supplementary Information

The online version contains supplementary material available at <https://doi.org/10.1186/s12974-021-02130-1>.

**Additional file 1: Supplement Figure 1.** Ratios of Corneal Infiltrating Cells to Nerve Fiber Parameters in Participants with Chronic Inflammatory Demyelinating Polyneuropathy (CIDP) or Diabetes and Effect of Glycemia on Corneal Infiltrating Cells. In participants with CIDP or diabetes and controls, ratios were calculated for (a–c) total corneal cell counts to corneal nerve fiber density (CNFD), corneal nerve branch density (CNBD), and corneal nerve fiber length (CNFL); (d–f) DCF + NCF (total number of infiltrating cells with proximity to nerve fibers) to CNFD, CNBD, and CNFL; and (g–i) DCP + NCP (total number of cells without nerve fiber contact) to CNFD, CNBD, and CNFL. All participants were stratified into two groups based on glycated hemoglobin (HbA<sub>1c</sub>) levels: >7.0% and ≤7.0% and corneal infiltrating cells were quantified (j–l). Mean ± SEM, \*P<0.05, \*\*P<0.01, \*\*\*P<0.001, ns indicates not significant.

## Acknowledgements

Not applicable

## Authors' contributions

All authors had full access to the study data and take responsibility for the integrity and accuracy of the data analysis. Study concept and design: Stettner, Hinrichs, Lee. Acquisition, analysis, or interpretation of data: Lee, Hinrichs, Petropoulos, Stettner, and Erdlenbruch. Drafting of the manuscript: Stettner and Fleischer. Critical revision of the manuscript for important intellectual content: Stettner, Malik, Hartung, Kleinschnitz, and Kieseier. Statistical analysis: Fleischer, Lee, Hinrichs, Stettner, and Petropoulos. Administrative, technical, or material support: Lee, Petropoulos, and Fleischer. Study supervision: Stettner. All authors read and approved the final manuscript.

## Funding

No funding was received for this study. Open Access funding enabled and organized by Projekt DEAL.

**Availability of data and materials**

The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

**Declarations****Ethics approval and consent to participate**

Statement on ethics approval and consent was included. The name of the ethics committee that approved the study and the committee's reference number was included.

**Consent for publication**

Not applicable

**Competing interests**

Fleischer (none), Lee (none), Hinrichs (none), Petropoulos (none), Erdlenbruch (none), Malik served on the scientific advisory board and/or received speaker honoraria or travel funding from Biogen Idec, Merck, Pfizer, Novo Nordisk. Hartung received personal fees for consulting, serving on steering, and data monitoring committees from Bayer Healthcare, Biogen, Celgene Receptos, CSL Behring, GeNeuro, MedImmune, Merck, Novartis, Octapharma, Roche, Sanofi Genzyme, and Teva. Kieseier (none), Kleinschnitz (none), Stettner: served on the scientific advisory and/or received speaker honoraria or travel funding from UCB, Biogen Idec, Grifols, Genzyme, Roche, Merck, Novartis, Octapharma, CSL Behring, Sanofi-Aventis, TEVA, and Bayer.

**Author details**

<sup>1</sup>Department of Neurology and Center for Translational and Behavioral Neurosciences (C-TNBS), University Hospital Essen, University Duisburg-Essen, Essen, Germany. <sup>2</sup>Department of Neurology, Medical Faculty, Heinrich-Heine-University, Düsseldorf, Germany. <sup>3</sup>Department of Cardiology, University Hospital Essen, University Duisburg-Essen, Essen, Germany. <sup>4</sup>Institute of Cardiovascular Science, Faculty of Medical and Human Sciences, University of Manchester, Manchester, UK. <sup>5</sup>Weill Cornell Medicine-Qatar, Educator City, Doha, Qatar. <sup>6</sup>Brain and Mind Centre, University of Sydney, Sydney, Australia. <sup>7</sup>Medical University Vienna, Vienna, Austria. <sup>8</sup>Department of Neurology, Palacky University, Olomouc, Czech Republic.

Received: 7 August 2020 Accepted: 15 March 2021

Published online: 08 April 2021

**References**

1. Querol L, Crabtree M, Herepath M, Priedane E, Viejto Viejto I, Agush S, Sommerer P. 2020. Systematic literature review of burden of illness in chronic inflammatory demyelinating polyneuropathy (CIDP). *J Neurol*
2. McLeod JG, Pollard JD, Macaskill P, Mohamed A, Spring P, Khurana V. Prevalence of chronic inflammatory demyelinating polyneuropathy in New South Wales, Australia. *Ann Neurol*. 1999;46(6):910–3. [https://doi.org/10.1002/1531-8249\(199912\)46:6<910::AID-ANA14>3.0.CO;2-2](https://doi.org/10.1002/1531-8249(199912)46:6<910::AID-ANA14>3.0.CO;2-2).
3. Chaudhary UJ, Rajabally YA. 2020. Underdiagnosis and diagnostic delay in chronic inflammatory demyelinating polyneuropathy. *J Neurol*. DOI: <https://doi.org/10.1007/s00415-020-10287-7>
4. Liberatore G, Manganelli F, Doneddu PE, Cocito D, Fazio R, Briani C, Filosto M, Benedetti L, Mazzeo A, Antonini G, Cosentino G, Jann S, Cortese A, Marfia GA, Clerici AM, Siciliano G, Carpo M, Luigetti M, Lauria G, Rosso T, Cavaletti G, Santoro L, Peci E, Tronci S, Ruiz M, Cotti Piccinelli S, Schenone A, Leonardi L, Toscano A, Mataluni G, Spina E, Gentile L, Nobile-Orazio E. 2020. Chronic inflammatory demyelinating polyradiculoneuropathy: can a diagnosis be made in patients not fulfilling electrodiagnostic criteria? *Eur J Neurol*
5. Stino AM, Naddaf E, Dyck PJ, Dyck PJB. 2020. Chronic inflammatory demyelinating polyradiculoneuropathy-Diagnostic pitfalls and treatment approach. *Muscle Nerve*
6. Rajabally YA, Stettner M, Kieseier BC, Hartung HP, Malik RA. CIDP and other inflammatory neuropathies in diabetes - diagnosis and management. *Nat Rev Neurol*. 2017;13(10):599–611. <https://doi.org/10.1038/nrneuro.2017.123>.
7. Doneddu PE, Cocito D, Manganelli F, Fazio R, Briani C, Filosto M, Benedetti L, Bianchi E, Jann S, Mazzeo A, Antonini G, Cosentino G, Marfia GA, Cortese A, Clerici AM, Carpo M, Schenone A, Siciliano G, Luigetti M, Lauria G, Rosso T, Cavaletti G, Beghi E, Liberatore G, Santoro L, Spina E, Peci E, Tronci S, Ruiz M, Cotti Piccinelli S, Verrengia EP, Gentile L, Leonardi L, Mataluni G, Piccolo L, Nobile-Orazio E. Frequency of diabetes and other comorbidities in chronic inflammatory demyelinating polyradiculoneuropathy and their impact on clinical presentation and response to therapy. *J Neurol Neurosurg Psychiatry*. 2020;91(10):1092–9. <https://doi.org/10.1136/jnnp-2020-0323615>.
8. Efron N, Edwards K, Roper N, Pritchard N, Sampson GP, Shahidi AM, Vagenas D, Russell A, Graham J, Dabbah MA, Malik RA. Repeatability of measuring corneal subbasal nerve fiber length in individuals with type 2 diabetes. *Eye Contact Lens*. 2010;36(5):245–8. <https://doi.org/10.1097/CL.0b013e3181ee915>.
9. Hertz P, Bril V, Orszag A, Ahmed A, Ng E, Nwe P, Ngo M, Perkins BA. Reproducibility of in vivo corneal confocal microscopy as a novel screening test for early diabetic sensorimotor polyneuropathy. *Diab Med*. 2011;28(10):1253–60. <https://doi.org/10.1111/j.1464-5491.2011.03299.x>.
10. Smith AG, Kim G, Porzio M, Allen B, Koach M, Miffilin M, Digre K, Keung BM, Singleton JR. Corneal confocal microscopy is efficient, well-tolerated, and reproducible. *J Peripher Nervous Syst*. 2013;18(1):54–8. <https://doi.org/10.1111/jns5.12008>.
11. Malik RA, Kallinikos P, Abbott CA, van Schie CH, Morgan P, Efron N, Boulton AJ. Corneal confocal microscopy: a non-invasive surrogate of nerve fibre damage and repair in diabetic patients. *Diabetologia*. 2003;46(5):683–8. <https://doi.org/10.1007/s00125-003-1086-8>.
12. Perkins BA, Lovblom LE, Bril V, Scarr D, Ostrovski I, Orszag A, Edwards K, Pritchard N, Russell A, Dehghani C, Pacaud D, Romanchuk K, Mah JK, Jeziorska M, Marshall A, Shtein RM, Pop-Busui R, Lentz SJ, Boulton AJM, Tavakoli M, Efron N, Malik RA. Corneal confocal microscopy for identification of diabetic sensorimotor polyneuropathy: a pooled multinational consortium study. *Diabetologia*. 2018;61(8):1856–61. <https://doi.org/10.1007/s00125-018-4653-8>.
13. Ferdousi M, Kalteniece A, Azmi S, Petropoulos IN, Ponirakis G, Alam U, Asghar O, Marshall A, Fullwood C, Jeziorska M, Abbott C, Lauria G, Faber CG, Soran H, Efron N, Boulton AJM, Malik RA. 2020. Diagnosis of neuropathy and risk factors for corneal nerve loss in type 1 and type 2 diabetes: a corneal confocal microscopy study. *Diabetes Care*
14. Chen X, Graham J, Dabbah MA, Petropoulos IN, Ponirakis G, Asghar O, Alam U, Marshall A, Fadavi H, Ferdousi M, Azmi S, Tavakoli M, Efron N, Jeziorska M, Malik RA. Small nerve fiber quantification in the diagnosis of diabetic sensorimotor polyneuropathy: comparing corneal confocal microscopy with intraepidermal nerve fiber density. *Diab Care*. 2015;38(6):1138–44. <https://doi.org/10.2337/dc14-2422>.
15. Pritchard N, Edwards K, Russell AW, Perkins BA, Malik RA, Efron N. Corneal confocal microscopy predicts 4-year incident peripheral neuropathy in type 1 diabetes. *Diab Care*. 2015;38(4):671–5. <https://doi.org/10.2337/dc14-2114>.
16. Tavakoli M, Marshall A, Pitceathly R, Fadavi H, Gow D, Roberts ME, Efron N, Boulton AJ, Malik RA. Corneal confocal microscopy: a novel means to detect nerve fibre damage in idiopathic small fibre neuropathy. *Exp Neurol*. 2010; 223(1):245–50. <https://doi.org/10.1016/j.jepneurol.2009.08.033>.
17. Tavakoli M, Marshall A, Banka S, Petropoulos IN, Fadavi H, Kingston H, Malik RA. Corneal confocal microscopy detects small-fiber neuropathy in Charcot-Marie-Tooth disease type 1A patients. *Muscle Nerve*. 2012;46(5):698–704. <https://doi.org/10.1002/mus.23377>.
18. Kemp HI, Petropoulos IN, Rice ASC, Vollert J, Maier C, Sturm D, Schargus M, Peto T, Hau S, Chopra R, Malik RA. Use of Corneal Confocal Microscopy to Evaluate Small Nerve Fibers in Patients With Human Immunodeficiency Virus. *JAMA Ophthalmol*. 2017;135(7):795–800. <https://doi.org/10.1001/jamaophthalmol.2017.1703>.
19. Ferdousi M, Azmi S, Petropoulos IN, Fadavi H, Ponirakis G, Marshall A, Tavakoli M, Malik I, Mansoor W, Malik RA. Corneal Confocal Microscopy Detects Small Fibre Neuropathy in Patients with Upper Gastrointestinal Cancer and Nerve Regeneration in Chemotherapy Induced Peripheral Neuropathy. *PLoS One*. 2015;10(10):e0139394. <https://doi.org/10.1371/journal.pone.0139394>.
20. Rousseau A, Cauquil C, Dupas B, Labbé A, Baudouin C, Barreau E, Théaudin M, Lacroix C, Guiochon-Mantel A, Benmalek A, Labetoulle M, Adams D. Potential role of in vivo confocal microscopy for imaging corneal nerves in transthyretin familial amyloid polyneuropathy. *JAMA Ophthalmol*. 2016;134: 983–9. <https://doi.org/10.1001/jamaophthalmol.2016.1889>.
21. Pagovich OE, Vo ML, Zhao Z, Petropoulos IN, Yuan M, Lertsuwanroj B, Ciralsky J, Lai E, Kiss S, D'Amico DJ, Mezey JG, Malik RA, Crystal RG. 2018. Corneal Confocal Microscopy: Neurologic Disease Biomarker in Friedreich's Ataxia. *Ann Neurol*

22. Leppin K, Behrendt AK, Reichard M, Stachs O, Guthoff RF, Baltrusch S, Eule JC, Vollmar B. Diabetes mellitus leads to accumulation of dendritic cells and nerve fiber damage of the subbasal nerve plexus in the cornea. *Invest Ophthalmol Vis Sci.* 2014;55(6):3603–15. <https://doi.org/10.1167/iovs.14-14307>.
23. Stettner M, Hinrichs L, Guthoff R, Bairo S, Petropoulos IN, Warnke C, Hartung HP, Malik RA, Kieseier BC. Corneal confocal microscopy in chronic inflammatory demyelinating polyneuropathy. *Ann Clin Transl Neurol.* 2016; 3(2):88–100. <https://doi.org/10.1002/acn3.275>.
24. Pitarokoiili K, Sturm D, Labedi A, Greiner T, Eitner L, Kumowski N, Enax-Krumova EK, Fisse AL, Maier C, Gold R, Tegenthoff M, Schmidt-Wilcke T, Yoon MS. Neuroimaging markers of clinical progression in chronic inflammatory demyelinating polyradiculoneuropathy. *Ther Adv Neurol Disord.* 2019;12:1756286419855485.
25. Athanasopoulos D, Motte J, Fisse AL, Grueter T, Trampe N, Sturm D, Tegenthoff M, Sgodzai M, Klimas R, Querol L, Gold R, Pitarokoiili K. Longitudinal study on nerve ultrasound and corneal confocal microscopy in NF155 paranodopathy. *Ann Clin Transl Neurol.* 2020;7(6):1061–8. <https://doi.org/10.1002/acn3.51061>.
26. Hughes RA, Bouche P, Cornblath DR, Evers E, Hadden RD, Hahn A, Illa I, Koski CL, Leger JM, Nobile-Orazio E, Pollard J, Sommer C, Van den Bergh P, van Doorn PA, van Schaik IN. European Federation of Neurological Societies/Peripheral Nerve Society guideline on management of chronic inflammatory demyelinating polyradiculoneuropathy: report of a joint task force of the European Federation of Neurological Societies and the Peripheral Nerve Society. *Eur J Neurol.* 2006;13(4):326–32. <https://doi.org/10.1111/j.1468-1331.2006.01278.x>.
27. Tesfaye S, Boulton AJ, Dyck PJ, Freeman R, Horowitz M, Kempner P, Lauria G, Malik RA, Spallone V, Vinik A, Bernardi L, Valensi P. Diabetic neuropathies: update on definitions, diagnostic criteria, estimation of severity, and treatments. *Diab Care.* 2010;33(10):2285–93. <https://doi.org/10.2337/dc10-1303>.
28. Rajabally YA, Peric S, Cobeljic M, Afzal S, Bozovic I, Palibrk A, Basta I. Chronic inflammatory demyelinating polyneuropathy associated with diabetes: a European multicentre comparative reappraisal. *J Neurol Neurosurg Psychiatry.* 2020;91(10):1100–4. <https://doi.org/10.1136/jnnp-2020-322971>.
29. Lotan I, Hellman MA, Steiner I. Diagnostic criteria of chronic inflammatory demyelinating polyneuropathy in diabetes mellitus. *Acta Neurol Scand.* 2015;132(4):278–83. <https://doi.org/10.1111/ane.12394>.
30. Dunnigan SK, Ebadi H, Breiner A, Katzberg HD, Lovblom LE, Perkins BA, Bril V. Conduction slowing in diabetic sensorimotor polyneuropathy. *Diab Care.* 2013;36(11):3684–90. <https://doi.org/10.2337/dc13-0746>.
31. Herrmann DN, Ferguson ML, Logigian EL. Conduction slowing in diabetic distal polyneuropathy. *Muscle Nerve.* 2002;26(2):232–7. <https://doi.org/10.1002/mus.10204>.
32. Dunnigan SK, Ebadi H, Breiner A, Katzberg HD, Lovblom LE, Perkins BA, Bril V. Comparison of diabetes patients with "demyelinating" diabetic sensorimotor polyneuropathy to those diagnosed with CIDP. *Brain Behav.* 2013;3(6):656–63. <https://doi.org/10.1002/brb3.177>.
33. Mitsuma S, Van den Bergh P, Rajabally YA, Van Parijs V, Martin-Lamb D, Sonoo M, Inaba A, Shimizu T, Iose S, Sato Y, Komori T, Misawa S, Kuwabara S. Effects of low frequency filtering on distal compound muscle action potential duration for diagnosis of CIDP: A Japanese-European multicenter prospective study. *Clin Neurophysiol.* 2015;126(9):1805–10. <https://doi.org/10.1016/j.clinph.2014.11.027>.
34. Rajabally YA, Martin-Lamb D, Nicolas G. Compound muscle action potential amplitude and distal potential duration in axonal neuropathy. *Muscle Nerve.* 2014;49(1):146–7. <https://doi.org/10.1002/mus.24065>.
35. Jann S, Bramero MA, Beretta S, Koch S, Defanti CA, Toyka KV, Sommer C. Diagnostic value of sural nerve matrix metalloproteinase-9 in diabetic patients with CIDP. *Neurology.* 2003;61(11):1607–10. <https://doi.org/10.1212/01.WNL.0000096174.6850.7F>.
36. Sommer C, Koch S, Lammens M, Gabreels-Festen A, Stoll G, Toyka KV. Macrophage clustering as a diagnostic marker in sural nerve biopsies of patients with CIDP. *Neurology.* 2005;65(12):1924–9. <https://doi.org/10.1212/01.wnl.0000188879.19900.b7>.
37. Mayer WJ, Mackert MJ, Kranebitter N, Messmer EM, Gruterich M, Kampik A, Kook D. Distribution of antigen presenting cells in the human cornea: correlation of in vivo confocal microscopy and immunohistochemistry in different pathologic entities. *Curr Eye Res.* 2012;37(11):1012–8. <https://doi.org/10.3109/02713683.2012.696172>.
38. Chen W, Hara K, Tian Q, Zhao K, Yoshitomi T. Existence of small slow-cycling Langerhans cells in the limbal basal epithelium that express ABCG2. *Exp Eye Res.* 2007;84(4):626–34. <https://doi.org/10.1016/j.exer.2006.11.006>.
39. Mayer WJ, Irschick UM, Moser P, Wurm M, Huemer HP, Romani N, Irschick EU. Characterization of antigen-presenting cells in fresh and cultured human corneas using novel dendritic cell markers. *Invest Ophthalmol Vis Sci.* 2007;48(10):4459–67. <https://doi.org/10.1167/iovs.06-1184>.
40. Alkawahjah NM, Dunnigan SK, Bril V. Comparison of monoclonal gammopathy of undetermined significance-associated neuropathy and chronic inflammatory demyelinating polyneuropathy patients. *J Neurol.* 2014;261(8):1485–91. <https://doi.org/10.1007/s00415-014-7357-0>.
41. Nabi S, Kahlon P, Bozorgnia F, Arshad A, Saleem A, Kuriakose P. Analyzing Relationship Between Monoclonal Gammopathy of Undetermined Significance (MGUS) with Different Types of Neuropathy: An Observational Study. *Indian J Hematol Blood Transfus.* 2016;32(2):186–92. <https://doi.org/10.1007/s12288-015-0547-9>.
42. Magy L, Chassande B, Maisonnobe T, Bouche P, Vallat JM, Leger JM. Polyneuropathy associated with IgG/IgA monoclonal gammopathy: a clinical and electrophysiological study of 15 cases. *Eur J Neurol.* 2003;10(6): 677–85. <https://doi.org/10.1046/j.1468-1331.2003.00687.x>.

## Publisher's Note

Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

**Ready to submit your research? Choose BMC and benefit from:**

- fast, convenient online submission
- thorough peer review by experienced researchers in your field
- rapid publication on acceptance
- support for research data, including large and complex data types
- gold Open Access which fosters wider collaboration and increased citations
- maximum visibility for your research: over 100M website views per year

**At BMC, research is always in progress.**

Learn more [biomedcentral.com/submissions](https://biomedcentral.com/submissions)

