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REVIEW

# The Therapeutic Potential of Dipeptidyl Peptidase 4 Inhibitors and Glucagon-Like Peptide-1 Receptor Agonists in Diabetic Peripheral Neuropathy

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## ABSTRACT

Diabetic peripheral neuropathy (DPN) is one of the commonest complications of diabetes mellitus (DM). Current therapeutic approaches largely focus on pain management. However, less evidence is available on the clinical potential of two widely prescribed drug categories in DM management: dipeptidyl peptidase 4 inhibitors (DPP-4is) and glucagon-like peptide-1 receptor agonists (GLP-1RAs). In this review, we discuss evidence from both experimental and clinical studies on the potential utility of these drugs in the management of DPN. Immunohistochemical data indicate that agents in both categories promote neurite outgrowth, ion conduction, neuronal survival and Schwann cell function. Furthermore, intra-epidermal nerve fibre density has been reported to increase with DPP-4is or GLP-1RAs treatment. Moreover, electrophysiological studies have indicated a diverse, but mostly beneficial, effect on motor or sensory nerve conduction velocity. Clinical tests, such as the muscular grip or paw jumping control resembling neuropathic symptoms, have also confirmed the advantageous effect of DPP-4is and GLP-1RAs. Finally, limited but promising clinical data have shown improved somatosensory-evoked potentials and vibration perception threshold, as well as restored excitability and nerve size parameters. Nevertheless, further clinical studies are required to elucidate the exact role of DPP-4is and GLP-1RAs in DPN.

**Keywords:** Type 1 diabetes mellitus; Type 2 diabetes mellitus; Diabetic peripheral neuropathy; Dipeptidyl peptidase 4 inhibitors; Glucagon-like peptide-1 receptor agonists

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| Key Summary Points |
| --- |
| *Why conduct this review?* |
| Diabetic peripheral neuropathy (DPN) is one of the most frequent complications of diabetes mellitus (DM) |
| Dipeptidyl peptidase 4 inhibitors (DPP-4is) and glucagon-like peptide-1 receptor agonists (GLP-1RAs) are two widely prescribed antidiabetic agents, but their role in DPN has not been widely studied |
| *What was learned from the review?* |
| Experimental data show increased neurite outgrowth and intra-epidermal nerve fibre density (IENFD) with DPP-4is or GLP-1RAs |
| Recent clinical data also encourage their use, but further large-scale studies are necessary for their introduction to clinical practice |

## INTRODUCTION

Diabetic peripheral neuropathy (DPN) is the most frequent microvascular complication of diabetes mellitus (DM) [1]. It may reduce protective sensation, lead to neuropathic pain and impair health-related quality of life (HRQoL) [2, 3]. Current treatment guidelines focus primarily on glucose control and pain alleviation [2]. However, less is known on the potential role of dipeptidyl peptidase 4 inhibitors (DPP-4is) and glucagon-like peptide-1 receptor agonists (GLP-1RAs) in DPN [4, 5]. Early data were not promising, but more recent evidence has sparked new interest in this therapeutic approach [6].

Both of these pharmacological categories enhance the incretin effect, which is seriously impaired in subjects with type 2 diabetes mellitus (T2DM) [7]. This effect is mediated by GLP-1 and glucose-dependent insulinotropic peptide (GIP), and it is enhanced by the inhibition of DPP-4 promoting GLP-1 degradation [7, 8]. In addition, GLP-1RAs in particular are gaining persistent attention for their pleiotropic actions beyond glucose control [9].

The aim of this review was to discuss the evidence from basic research and clinical studies on the potential utility of these drugs in DPN. The review is based on previously conducted studies and does not contain any new studies with human participants or animals performed by any of the authors.

## SEARCH STRATEGY

We searched Scopus, PubMed/MEDLINE and Google Scholar databases for articles without time restriction, using combinations of the following key words: “diabetic neuropathy”, “polyneuropathy” “diabetic peripheral neuropathy”, “distal symmetrical polyneuropathy”, “type 1 diabetes mellitus”, “type 2 diabetes”, “glucagon-like peptide-1 receptor agonists” and “dipeptidyl peptidase 4 inhibitors”. All types of articles (clinical trials, meta-analyses, case–control studies, observational studies, cross-sectional studies, prospective/retrospective studies, cohort studies, comparative studies, randomised controlled studies, experimental studies) were included. Articles on autonomic neuropathy were excluded. Only articles in English were considered.

## EXPERIMENTAL STUDIES WITH GLP-1RAS IN DPN

Experimental studies on GLP-IRAs in the management of DPN are summarised in Table 1.

GLP-1RAs have been extensively studied in DPN in various animal models and cell lines. Early evidence from 2008 denoted the importance of GLP-1 in the nervous system: GLP-1 administration protected nerve cells from degeneration or death under the condition of simultaneous deprivation of nerve growth factor (NGF), a major neurotrophic factor mediating the survival of neurons [10]. GLP-1 suppressed the induction of Bim, a pro-apoptotic protein whose induction is promoted under NGF deprivation

**Table 1** Experimental studies on GLP-1RAs in DPN

| Study | Animal/experimental model and study design | Major outcomes | Conclusions |
|---|---|---|---|
| Kan et al. [13] | STZ-induced T1DM mice and T2DM db/db mice (diabetes duration of 2 months and simultaneous neuropathy) treated with exendin-4 or different doses of insulin and immunohistochemical analysis | GLP-1 expression was observed in the dorsal root ganglia of diabetic and non-diabetic mice<br>In vitro administration of exendin-4 resulted in neurite outgrowth of adult sensory neurons<br>In T1DM, high-dose insulin administration restored glycaemic control, alleviated (partially) thermal sensory loss and increased intra-epidermal innervation without electophysiological improvement, while low-dose insulin was ineffective<br>Exendin-4 improved electrophysiological parameters and sensory perception<br>In T2DM, no effects were documented in hyperglycaemia, electrophysiolgical, behavioural indices or epidermal axon loss with insulin or exendin-4 administration<br>Exendin-4 administration resulted in motor electrophysiology improvement<br>RAGE or NF-κB neuronal expression was not affected by either diabetes or treatment | GLP-1RAs cannot fully reverse DPN |
| Kornelius et al. [24] | RSC96 cell line study (Schwann cells) exposed to diabetic mimicking conditions (high glucose and high free fatty acid) treated with liraglutide | Diabetes-mimicking conditions reduced viability of RSC96 Schwann cells by 51%<br>Liraglutide administration resulted in reduced oxidative stress levels by increasing the activity of antioxidant enzymes (SOD1/2 and catalase)<br>NF-κB-associated inflammation was inhibited by liraglutide<br>Liraglutide administration enhanced the expression of neurotrophic factors and myelination-related proteins which are linked with insulin signalling | Schwann cell dysfunction is attenuated by liraglutide; GLP-1 signalling modulation emerges promising |

**Table 1** continued

| Study | Animal/experimental model and study design | Major outcomes | Conclusions |
|---|---|---|---|
| Pandey et al. [17] | Cell line study featuring SH-SY5Y neuroblastoma cells treated with exendin-4 | Under high glucose conditions, exendin-4 treatment resulted in increased expression of survival markers pAKT/AKT and Bcl-2, decreased expression of the pro-apoptotic marker Bax and reduced expression of ROS defense markers (catalase, SOD-2 and HO-1)<br>Mitochondrial function analysis showed decreased expression of mitochondrial function-associated genes MCU and UCP3 and mitochondrial fission genes DRP1 and FIS1<br>Increased expression of mitochondrial homeostasis regulators Parkin and PINK1 following exendin-4 treatment was observed<br>Epac and AKT blockade inhibited the neuroprotective impact of exendin-4 treatment | GLP1-RAs exhibit neuroprotective actions by counteracting oxidative stress and mitochondrial dysfunction and by promoting neuronal survival through the Epac/AKT pathway |
| Himeno et al. [14] | C57BL6/J STZ-induced diabetic mice | GLP-1R expression in the dorsal root ganglion was confirmed<br>GLP-1 (7–37) and exendin-4 significantly enhanced neurite outgrowth<br>Both GLP-1RAs accelerated impaired neurite outgrowth of the neurons in the dorsal root ganglion cultured with Schwann cell-conditioned media under diabetes mimicking conditions<br>Exendin-4 administration at the dose used had no impact on glycaemic control<br>Neuropathic symptoms, such as hypoalgesia and electrophysiological parameters (MNCV, SNCV) were improved with exendin-4 treatment without decreasing SNBF<br>Decreased IENFD was attenuated with exendin-4 treatment | Exendin-4 holds therapeutic potential for DPN |
| Komsuoglu Celikyurt et al. [21] | BALB/c STZ-induced diabetic mice treated with exenatide | In the hotplate test, latency of mice to lick hindpaws was significantly increased ($p < 0.01$), indicative of impaired pain reaction<br>Exenatide treatment resulted in decreased latency, indicative of improved reaction ($p < 0.05$) | Exenatide may serve as a promising therapeutic approach |

**Table 1** continued

| Study | Animal/experimental model and study design | Major outcomes | Conclusions |
|---|---|---|---|
| Jolivalt et al. [22] | STZ-induced diabetic rats treated with exenatide | GLP-1R expression was observed in rat dorsal root ganglia, both in neurons and in Schwann cells<br>Diabetic conditions did not change protein levels<br>GLP-1RAs actions were not mediated by ERK1/2 in sciatic nerve of control rats<br>GLP1-RAs administration resulted in significantly increased pERK1/2 in sciatic nerves of diabetic rats, with increased function ($p < 0.01$), which was inhibited by exendin 9–39 ($p < 0.05$)<br>Exenatide administration led to significantly increased GLP-1R expression ($p < 0.01$), but it did not affect glycaemic control, insulin levels and temperature perception (thermal response latencies)<br>Decreased MNCV ($p < 0.05$,after 1 month and $p < 0.01$ at the end of the study) and IENFD ($p < 0.05$) were ameliorated by exenatide administration | GLP-1R-mediated ERK-signalling pathway may protect large motor fibre and small C-fibre structure independently of glycaemic control |
| Luciani et al. [16] | Cell line study featuring SH-SY5Y neuroblastoma cells | GLP-1R was expressed in SH-SY5Y cell line<br>Exendin-4 counteracted oxidative stress and $H_2O_2$-mediated cell apoptosis<br>Exendin-4 administration resulted in increased number of neurites, mediated through intracellular actin and tubulin changes<br>Electrophysiological analysis indicated increased cell membrane surface, increased $Na^+$ conduction, increased $Ca^{2+}$ currents (both T- and L-type), overall indicative of a mature neuronal phenotype | Exendin-4 promotes neuronal differentiation and may serve as a therapeutic approach |

**Table 1** continued

| Study | Animal/experimental model and study design | Major outcomes | Conclusions |
|---|---|---|---|
| Liu et al. [15] | Diabetic and non-diabetic rats treated with either exendin-4 or placebo (over a study period of 24 weeks) | GLP-1 expression was observed in the sciatic nerve and skin<br>Exendin-4 treatment resulted in reduced current perception thresholds at 2000 Hz at weeks 16 and 24 (both $p < 0.05$) and 250 Hz in diabetic rats at week 24 ($p < 0.05$)<br>The decrease in myelinated fibre size ($p < 0.05$), decreased axon/fibre area ratio in the sciatic nerve ($p < 0.05$) and loss of IENFD at week 16 and 24 ($p < 0.05$ and $p < 0.01$, respectively) were attenuated by exendin-4 administration<br>Significantly increased nerve fibre density of the hind dorsum was observed with exendin-4 treatment ($p < 0.05$)<br>Compared with controls, Schwann cell apoptosis was diminished and cAMP levels increased (both $p < 0.05$) | Exendin-4 ameliorated peripheral nerve degeneration |
| Tsukamoto et al. [20] | 3-month-old Wistar rat dorsal root ganglion samples and PC12 cells treated with exendin-4 | GLP-1R expression was mainly observed in small and large peptidergic neurons<br>Increased neurite outgrowth and neuronal survival were observed with exendin-4 in a dose-dependent way, at 2 and 7 days in culture, respectively<br>Treatment with the highest dose (100 nM) resulted in increased neurite outgrowth and viability of neurons in dorsal root ganglion<br>Suppression of RhoA, an inhibitory regulator of peripheral nerve regeneration, resulted also from the treatment under the highest exendin-4 dose in PC12 cells<br>These effects on neurite outgrowth and neuronal survival were further enhanced by the co-administration of exendin-4 and LY294002, a PI3K inhibitor | Exendin-4 exerts its actions through the PI3K pathway, which suppresses RhoA activity; it may compensate decreased insulin impact on neurons in DPN |

**Table 1** continued

| Study | Animal/experimental model and study design | Major outcomes | Conclusions |
|---|---|---|---|
| Mohiuddin et al. [18] | Cell line study of neuronal 50B11 cells treated with either exendin-4 or GLP-1 (7–37) | MTS assay showed that GLP-1RAs ameliorated decreased cell viability caused by oxidative stress<br>Both agents applied were not cytotoxic<br>GLP-1RAs administration resulted in SOD activation, indicative of increased antioxidant enzyme activity<br>No effects on neuronal markers were observed with GLP-1RAs treatment<br>Exendin-4 treatment counteracted oxidative stress-driven apoptosis<br>GLP-1 RAs administration inhibited cell proliferation inhibition and increased neurite projections<br>Increased cAMP levels were not observed | GLP-1 RAs show neuroprotective effects and may be used as therapeutic agents in DPN |
| Biswas et al. [10] | Neuronal PC12 cells treated with GLP-1 | GLP-1 administration resulted in persistent protection of cells from degeneration or death under NGF deprivation<br>GLP-1 inhibited Bim induction, a pro-apoptotic protein which is promoted by NGF deprivation | GLP-1 action as a neuroprotective agent is partially mediated by Bim inhibition |
| Perry et al. [19] | Neuronal PC12 cells treated with exendin-4 and exendin-4 WOT | GLP-1 and exendin-4 administration resulted in neurite outgrowth, resembling the action of NGF<br>Co-incubation with exendin (9–39) mitigated this effect<br>All effects were noted in the absence of cytotoxic conditions or cellular dysfunction | GLP-1 emerges as a key mediator with neuroprotective potential in DPN and neurodegenerative diseases |

**Table 1** continued

| Study | Animal/experimental model and study design | Major outcomes | Conclusions |
|---|---|---|---|
| Fontanella et al. [28] | Neuroblastoma cell line (SH-SY5Y) exposed to high glucose and treated with tirzepatide | Significantly reduced Ki-67, indicative of cell proliferation, was observed under concomitant high glucose levels and with tirzepatide administration ($p < 0.05$)<br>Significantly increased CREB and BDNF expression (both mRNA and protein levels) were observed ($p < 0.05$)<br>Significantly increased Bcl-2 and significantly decreased Bax protein levels, without any effect on mRNA levels, were documented with tirzepatide treatment ($p < 0.05$), but without any effects under concomitant high glucose levels<br>Tirzepatide administration resulted in significantly elevated pAKT, MAP2, GAP43, and AGBL4 and in significantly increased GAP43 and AGBL4, indicative of neuronal cell differentiation;<br>AGBL4 protein and mRNA levels were not affected under concomitant high glucose levels<br>Significantly elevated GLUT3 and GLUT4 mRNA and GLUT4 and SORBS1 were observed with tirzepatide treatment and also under high glucose levels<br>Tirzepatide administration counteracted high glucose-mediated increased DNA methylation of CREB and BDNF; no effects on DNA methylation of AGBL4 and SORBS1 were observed<br>Tirzepatide treatment resulted in significantly decreased miR-34a and increased miR-212 and miR-29c; the opposite effects were observed under high glucose conditions and were reversed by tirzepatide | Tirzepatide attenuated neurodegeneration under high-glucose and may serve as a therapeutic agent in DPN |

*AGBL4* Adenosine triphosphate/guanosine-5'-triphosphate binding protein like 4, *BDNF* brain-derived neurotrophic factor, *C57BL6/J* C57 black 6, *cAMP* cyclic adenosine monophosphate, *CREB* cyclic adenosine monophosphate response element-binding protein, *db/db* genetically diabetic-obese, *DPN* diabetic peripheral neuropathy, *DRP1* dynamin-related protein 1, *E3* ubiquitin-protein ligase, *Epac* exchange proteins directly activated by cAMP, *ERK* extracellular signal-regulated kinase, *FIS1* mitochondrial adaptor fission 1, *GAP43* growth-associated protein 43, *GLP-1* glucagon-like peptide-1, *GLP-1R* glucagon-like peptide-1 receptor, *GLP-1RAs* glucagon-like peptide 1 receptor agonists, *GLUT* glucose transporter, *HO-1* heme oxygenase-1, *IENFD* intraepidermal nerve fiber density, *L-type* long-lasting, *MAP2* microtubule-associated protein 2, *MCU* mitochondrial $Ca^{2+}$ uniport, *miR* micro RNA, *MNCV* motor nerve conduction velocity, *mRNA* messenger RNA, *MTS* 3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium inner salt, *NF-κB* nuclear factor kappa-light-chain-enhancer of activated B cells, *NGF* nerve growth factor, *pAKT* phosphorylated protein kinase B, *AKT* protein kinase B, *PI3K* phosphoinositide 3-kinase, *PINK1* phosphatase and tensin homolog-induced kinase 1, *RAGE* receptor for advanced glycated end products, *RhoA* Ras homolog family member A, *ROS* reactive oxygen species, *SNCV* sensory nerve conduction velocity, *SOD* superoxide dismutase, *SORBS1* sorbin and SH3 domain containing 1, *STZ* streptozocin, *T-type* transient opening, *T1DM* type 1 diabetes mellitus, *T2DM* type 2 diabetes mellitus, *UCP* uncoupling protein, *SNBF* sciatic nerve blood flow

conditions [10]. Consequently, GLP-1-mediated effects are partially promoted by Bim inhibition [10].

Exenatide, also termed exendin-4, has been extensively evaluated [11]. Exendin-4 has a short half-time and is considered to be relatively more resistant to degradation by dipeptidyl dipeptase-4 (DPP-4) [12]. In an experimental study using models of both T2DM and type 1 diabetes mellitus (T1DM), Kan et al. [13] confirmed that GLP-1 expression was present not only in the dorsal root ganglion of diabetic mice, but also in those of non-diabetic mice. In streptozocin (STZ)-induced T1DM mice, the administration of high-dose insulin resulted in optimal glucose control, reversed partially thermal sensory defects and increased intra-epidermal innervation without yielding any electrophysiological improvement, while low-dose insulin was ineffective [13]. In the same study, exendin-4 improved electrophysiological parameters and sensation and both insulin and exendin-4 administration had an insignificant impact on hyperglycaemia, electrophysiological indices or epidermal axon loss in T2DM db/db mice [13]. Interestingly, in T1DM mice these same authors reported a significant improvement of sensory nerve conduction velocity (SNCV) and a trend towards improved motor nerve condution velocity (MNCV), whereas the exact opposite effect was found in the T2DM model [13].

In a further experiment with another animal model, that of C57BL6/J STZ-induced diabetic mice [14], GLP-1R was expressed in the dorsal root ganglion, neurite outgrowth was improved by exendin-4 and exendin-4 treatment improved neuropathic symptoms (e.g. hypoalgesia), MNCV and SNCV, as well as intra-epidermal nerve fibre density (IENFD).

Diabetic and non-diabetic rats were treated with either exendin-4 or placebo in a 24-week study [15]. In diabetic rats, exendin-4 significantly improved current perception thresholds at 2000 Hz at week 16 ($p<0.05$); at week 24, this effect was significant at both 250 Hz and 2000 Hz (both $p<0.05$). Major improvements were noted in other nerve functional parameters upon exendin-4 administration: the initially observed decrease in myelinated fibre size ($p<0.05$), decreased axon/fibre area ratio in the sciatic nerve ($p<0.05$) and reduced IENFD, as evaluated by anti-rat protein gene product (PGP) 9.5, anti-GLP-1R (glucagon-like peptide 1 receptor) or anticaspase 3 at weeks 16 and 24 ($p<0.05$ and $p<0.01$, respectively), were ameliorated [15]. In this same study, exendin-4 significantly increased nerve fibre density of the hind dorsum ($p<0.05$), significantly decreased Schwann cell apoptosis ($p<0.05$) and increased cyclic adenosine monophosphate (cAMP) levels ($p<0.05$) [15].

The neuroblastoma-derived cell-line SH-SY5Y has also been studied [16, 17]. In the study of Luciani et al. [16], GLP-1R was found to be expressed in the SH-SY5Y cell-line. Oxidative stress and $H_2O_2$-mediated cell apoptosis were ameliorated by exendin-4, and the increased number of neurites upon exendin-4 administration was mediated through intracellular actin and tubulin changes [16]. These researchers focused on cell membrane electrophysiology and observed an increased cell membrane surface, increased $Na^+$ conduction through the respective channels and increased $Ca^{2+}$ currents (both T-[transient opening] and L-[long-lasting] type), features corresponding to a mature neuronal phenotype [16]. The recent study of Pandey et al.[17] focused on apoptosis-related indices and mitochondrial function in cells treated with exendin-4. The increased expression of survival markers (phosphorylated-AKT[pAKT]/protein kinase B [AKT] and Bcl-2) and decreased expression of pro-apoptotic marker Bax and reactive oxygen species (ROS) defence marker levels (catalase, superoxide dismutase 2 [SOD-2] and heme oxygenase-1 [HO-1]) were found following exendin-4 administration in high-glucose conditions [17]. Exendin-4 treatment had a major effect on mitochondrial function, as evidenced by the diminished expression of genes associated with mitochondrial function (mitochondrial $Ca^{2+}$ uniport [MCU] and uncoupling protein 3 [UCP3]) and mitochondrial fission genes (dynamin-related protein 1 [DRP1] and mitochondrial adaptor fission 1 [FIS1]), as well as by increased expression of mitochondrial homeostasis regulators, such as the RBR (RING-Between RING-RING) E3 ubiquitin-protein ligase Parkin and the phosphatase and tensin homolog-induced kinase 1 (PINK1) [17]. These

neuroprotective actions promoting survival were mediated by the Epac/AKT pathway [17].

The impact of GLP-1RAs (exendin-4 or GLP-1 (7–37)) on oxidative stress and neuronal function overall was discussed in a study of neuronal 50B11 cells [18]. In this study, the 3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium inner salt (MTS) assay was used to evaluate cell viability, with the results showing that GLP-1RAs attenuated oxidative stress-driven diminished viability and, in particular, exendin-4 oxidative stress-driven apoptosis. The administration of GLP-1RAs also led to SOD activation, corresponding to increased antioxidant enzyme activity, suppressed cell proliferation inhibition and facilitated neurite formation [18].

The neuronal PC12 cell line, a cell line that expresses the GLP-1 receptor, has also been studied [19]. GLP-1 and exendin-4 administration accelerated neurite outgrowth, an action which resembled that of NGF. Coincubation with the selective GLP-1R antagonist exendin (9–39) reversed this effect [19].

Tsukamoto et al. [20] analysed the effect of exendin-4 in the rat dorsal root ganglion of 3-month-old Wistar rats and also in PC12 cells. In both the in vitro and in vivo analyses, these authors observed that small and large peptidergic neurons expressed GLP-1R. Exendin-4 treatment increased neurite outgrowth and neuronal survival; this effect was dose-dependent and was seen at days 2 and 7 of culture. In PC12 cells, the highest exendin-4 dose (100 nM) resulted in the suppression of RhoA, an inhibitory regulator of peripheral nerve regeneration suppression [20]. These effects on neurite outgrowth and neuronal survival were enhanced by the coadministration of exendin-4 and LY294002, a phosphoinositide 3-kinase (PI3K) inhibitor [20].

Komsuoglu Celikyurt et al. [21] performed pain perception assays in BALB/c STZ-induced diabetic mice. In the hotplate test, the latency of the mice to lick the hindpaws was significantly increased ($p<0.01$), indicative of impaired pain reaction. Decreased latency was observed for exenatide-treated mice, indicative of improved reaction ($p<0.05$) [21].

STZ-induced diabetic rats were also assessed in relation to exenatide response [22]. In this study, the administration of GLP1-RAs resulted in significantly increased pERK1/2 levels in the sciatic nerves of diabetic rats, indicating increased function ($p<0.01$); these increased levels were suppressed by exendin 9–39 ($p<0.05$) [22]. The latter is known to inhibit GLP-1 actions [23]. Exenatide administration led to significantly increased GLP-1R expression in sciatic nerve samples from diabetic mice ($p<0.01$) [22]. MNCV ($p<0.05$ after 1 month; $p<0.01$ at the end of the study period) and IENFD ($p<0.05$) were significantly decreased in skin sections, but were normalised by exenatide [22].

Kornelius et al. [24] studied oxidative stress and inflammation in the RSC96 cell line (Schwann cells) exposed to high glucose and high free fatty acids and found that the viability of the RSC96 Schwann cells decreased by 51% under these conditions. Liraglutide administration reduced oxidative stress by increasing the activity of antioxidant enzymes, such as SOD1/2; it also suppressed the activity of nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB), thus reducing inflammation [24]. Liraglutide administration restored the expression of ciliary neurotrophic factor (CNTF), NGF, neurotrophin-3 (NT-3) and brain-derived neurotrophic factor (BDNF) [24]. Such neurotrophic factors are increasingly being considered as therapeutic agents in DPN [25, 26]. Of note, Gumuslu et al. [27] also observed the association between NGF and GLP-1R expression in the central nervous system. These authors reported reduced GLP-1R and NGF expression in the hippocampus of STZ/nicotinamide-induced diabetic BALB/cByJ mice. Following exenatide administration twice daily, the same authors observed a doubling of the expression of hippocampal GLP-1R and NGF compared to baseline over a period of 2 weeks [27]. Furthermore, notable improvement was found in myelination patterns: the mRNA levels of myelination-related proteins, such as proteolipid protein (PLP), myelin basic protein (MBP) and myelin protein zero (MPZ), were also found to be elevated, which are overall linked to insulin signalling [24]. The same analysis yielded restored receptor for advanced glycated end products (RAGE) expression following liraglutide administration, as an improvement was noted in the markedly elevated mRNA levels under the

diabetic conditions [24]. Interestingly, LY294002 reversed the liraglutide-induced effects in an insulin-AKT signalling-dependent manner [24].

A very recent study by Fontanella et al. [28] provided an insight into the potential effect of tirzepatide in DPN by studying the neuroblastoma cell line SH-SY5Y. Tirzepatide is termed a twincretin due to its role as a dual agonist of both GLP-1 and GIP receptors [29]. In Fontanella et al.'s study [28], a significantly decreased Ki-67 index was observed with tirzepatide administration under high-glucose levels, indicative of cell proliferation ($p < 0.05$). Substantially elevated BDNF expression (both mRNA and protein levels) was also noted ($p < 0.05$) by these authors. Tirzepatide also inhibited the apoptosis of neurons, increased Bcl-2 and decreased Bax protein levels, without any effect on mRNA levels ($p < 0.05$); these effects were mitigated under high-glucose levels. Tirzepatide treatment resulted in significantly elevated pAKT, microtubule-associated protein 2 (MAP2), growth associated protein 43 (GAP43) and adenosine triphosphate/guanosine-5'-triphosphate binding protein like 4 (AGBL4) protein and significantly increased GAP43 and AGBL4, indicative of neuronal cell differentiation; likewise, AGBL4 protein and mRNA levels were unaffected under concomitant high glucose. Significantly elevated GLUT3 (glucose transporter [GLUT] 3) and GLUT4 mRNA levels and significantly elevated GLUT4 and sorbin and SH3 domain containing 1 (SORBS1) protein levels, a major mediator of insulin signalling pathways and glucose uptake, were observed with tirzepatide treatment under high-glucose levels. Tirzepatide administration reversed high glucose-mediated increased DNA methylation of cyclic adenosine monophosphate response element-binding protein (CREB) and BDNF, without any effects on the methylation status of AGBL4 and SORBS1. Of note, tirzepatide treatment resulted in significantly decreased miR-34a (miR [microRNA]) (involved in neuronal growth) and increased levels of miR-212 (involved in apoptosis) and miR-29c (involved in differentiation), whereas the exact opposite effects were observed under high-glucose levels and these were efficiently reversed by tirzepatide [28].

## CLINICAL STUDIES WITH GLP-1RAS IN DPN

Clinical studies on GLP-IRAs in the management of DPN are summarised in Table 2.

Senyigit et al. [30] included T2DM subjects with and without microvascular diabetic complications (25 study participants with neuropathy) and 26 healthy participants. Significantly diminished DPP-4 ($p < 0.005$) and GLP-1 ($p < 0.001$) levels were seen in T2DM subjects. Interestingly, significantly elevated DPP-4 ($p < 0.05$) and total GLP-1 ($p < 0.001$) levels were present in T2DM subjects with complications. There was a positive correlation between DPP-4 levels and total GLP-1 ($r = 0.290$, $p < 0.01$), thus showing that DPP-4 activity cannot be used as a predictive marker of endogenous GLP-1 degradation. Among subjects with neuropathy, significantly elevated levels of DPP-4 ($p < 0.005$), total ($p < 0.001$) and active GLP-1 ($p < 0.01$) and secreted frizzled-related protein-4 (SFRP-4) ($p < 0.005$), were observed, compared to T2DM subjects without complications [30]. SFRP-4 is regarded as an adipokine capable of inducing insulin resistance [31].

In a recent observational study by Dhanapalaratnam et al. [32], 22 subjects with T2DM were treated either with semaglutide or dulaglutide. Before the intervention, increased nerve size was observed in 81.8% of participants; at 1 month after treatment, a significantly improved nerve size (decreased mean tibial nerve cross-sectional area [CSA]) was observed in 86.4% of participants ($p < 0.05$), while 31.8% showed normal nerve features ($< 12.4$ mm$^2$). CSA has been validated as a reliable ultrasonographic index of DPN [33]. The Total Neuropathy Score (TNS) of study participants was significantly reduced ($p = 0.039$) [32]. At 3 months, additional improvement was observed in 92.8% of participants ($p < 0.05$), along with a significant improvement in sural nerve conduction amplitude ($p < 0.05$); conversely, sural nerve conduction velocity was insignificantly affected at the 3-month follow-up ($p = 0.214$). At 3 months, the reductions in TNS persisted ($p = 0.025$), while significantly decreased modified Toronto Clinical

**Table 2** Clinical studies on GLP-1RAs in DPN

| Study | Study type | Study population and design | Major outcomes | Conclusions |
|---|---|---|---|---|
| Dhanapalaratnam et al. [32] | Observational study | 22 T2DM subjects treated with semaglutide or dulaglutide | Before treatment initiation, increased nerve size was observed in 81.8% of participants<br>At 1 month, 86.4% of participants exhibited improved nerve size (decreased mean tibial CSA) ($p < 0.05$), while 31.8% had normal nerve features (< 12.4 $mm^2$)<br>Significantly reduced TNS was observed ($p = 0.039$)<br>Insignificant reduction in mTCNS was documented ($p = 0.162$)<br>At 3 months, additional improvement was noted in 92.8% of participants ($p < 0.05$) along with improved sural nerve conduction amplitude ($p < 0.05$)<br>Sural NCV was not affected at 3 months ($p = 0.214$)<br>At 3 months, significantly reduced TNS was maintained ($p = 0.025$) and there was significant improvement in mTCNS ($p = 0.049$)<br>In a subgroup of 14 participants, continuous improvement of nerve CSA was observed at both study points ($p = 0.04$ and $p = 0.08$, respectively)<br>Dulaglutide was associated with insignificantly greater reduction in tibial CSA compared with semaglutide at both study points ($p = 0.464$ and $p = 0.454$, respectively) | GLP-1RAs resulted in improved nerve features and neurophysiological parameters |
| Jaiswal et al. [35] | 18-month proof-of-concept open-label randomised study | 46 T2DM subjects with mean age of 54 years, mean T2DM duration of 8 years and mean HbA1c at 8.2%,<br>22 participants<br>treated with exenatide (twice daily), and<br>24 study participants<br>treated with insulin glargine (daily) | DPN symptoms were present in 21 (96%) and 22 (92%) in the 2 treatment arms, respectively<br>Similar glycaemic control was observed between the two study arms ($p = 0.3$)<br>Insignificant differences were observed in the prevalence of confirmed clinical DPN, IENFD, NCV or QoL<br>Increased regeneration rate was observed only in the study arm treated with glargine ($p = 0.002$) compared with exenatide ($p = 0.06$) | 18-month exenatide treatment was not associated with changes in mild-to-moderate DPN |
| Dhanapalaratnam et al. [34] | Observational study | 14 participants treated with GLP-1RAs, and<br>Previously obtained data from 10 participants treated with exenatide | Significantly improved TNS was seen after 3-month GLP-1RAs ($p = 0.005$)<br>Sural amplitude was significantly increased ($p = 0.013$)<br>Improved $Na^+/K^+$ -ATPase pump function and $Na^+$ permeability were assessed following the study intervention | GLP-1RAs hold therapeutic potential for DPN |

**Table 2** continued

| Study | Study type | Study population and design | Major outcomes | Conclusions |
|---|---|---|---|---|
| Issar et al. [36] | Comparative study | 32 participants treated with exenatide<br>31 participants treated with DPP-4is<br>27 participants treated with SGLT-2is,<br>32 healthy individuals, and<br>10 participants evaluated prospectively for 3 months under exenatide treatment | DPP-4is therapy was not associated with optimal nerve excitability, as abnormal peak threshold reduction ($p < 0.001$), S2 accommodation ($p < 0.001$), superexcitability ($p < 0.001$), and subexcitability ($p < 0.001$) were observed<br>SGLT-2is therapy was not associated with significant improvement, as abnormal peak depolarisation threshold reduction ($p = 0.047$), S2 accommodation ($p < 0.01$), superexcitability ($p < 0.001$), and subexcitability ($p = 0.049$) were observed<br>Subjects treated with exenatide exhibited normal nerve excitability, both in the<br>Subjects in the prospective study arm treated with exenatide showed increased S2 accommodation ($p = 0.028$), increased superexcitability ($p = 0.037$) and decreased subexcitability ($p = 0.045$), independently of glycaemic control<br>Nerve excitability changes were not correlated with motor item of TNS | Peripheral nerve function is improved by exenatide |
| Senyigit et al. [30] | Observational study | T2DM subjects with and without microvascular complications (25 participants with DPN) and 26 healthy participants | Significantly decreased DPP-4 ($p < 0.005$) and GLP-1 ($p < 0.001$) levels were observed in T2DM subjects<br>CLU, amylin and active GLP-1 were similar between T2DM subjects and the control arm<br>T2DM subjects with complications showed significantly elevated DPP-4 ($p < 0.05$) and total GLP-1 ($p < 0.001$)<br>There was a positive correlation between DPP-4 and total GLP-1 ($r = 0.290$, $p < 0.01$)<br>Among subjects with DPN, significantly higher DPP-4 ($p < 0.005$), SfRP-4 ($p < 0.005$), total GLP-1 ($p < 0.001$) and active GLP-1 ($p < 0.01$) were observed, compared with T2DM subjects without complications | DPP-4 and GLP-1 (both active and total) were elevated in T2DM subjects with DPN |

*T2DM* type 2 diabetes mellitus, *GLP-1RAs* glucagon-like peptide 1 receptor agonists, *GLP-1* glucagon-like peptide 1, *CSA* nerve cross-sectional area, *TNS* total neuropathy score, *mTCNS* modified Toronto clinical neuropathy scale, *NCV* nerve conduction velocity, DPN diabetic peripheral neuropathy, *HbA1c* glycated haemoglobin, *IENFD* intra-epidermal nerve fiber density, *QoL* quality of life, *$Na^+/K^+$-ATPase* sodium–potassium adenosine triphosphatase, *DPP-4is* dipeptidyl peptidase 4 inhibitors, *DPP-4* dipeptidyl peptidase-4, *SGLT-2is* sodium-glucose cotransporter 2 inhibitors, *CLU* serum clusterin, *SFRP-4* secreted frizzled-related protein-4

Neuropathy Scale (mTCNS) score was also improved ($p=0.049$) [32]. In a subgroup of 14 participants, there was continuous significant improvement of nerve CSA at both study points ($p=0.04$ and $p=0.08$, respectively) [32]. Dulaglutide treatment was associated with an insignificantly greater reduction in tibial CSA compared with semaglutide at both study points ($p=0.464$ and $p=0.454$, respectively) [32].

A subsequent study by Dhanapalaratnam et al. [34] included 14 participants treated with GLP-1RAs and previously obtained data from 10 study participants treated with exenatide. Significantly improved TNS was found after 3 months of GLP-1RA therapy ($p=0.005$) [34]. Sural amplitude was also significantly increased ($p=0.013$) [34].

In an 18-month proof-of-concept open-label randomised study, 46 T2DM subjects were allocated either to the exenatide treatment arm (22 participants) or to the insulin glargine treatment arm (24 participants) [35]. DPN symptoms were observed in 21 (96%) subjects in the exenatide treatment arm and in 22 (92%) subjects in the insulin glargine treatment arm. No differences in glycaemic control were observed between the two study arms ($p=0.3$). Exenatide was not associated with any benefit in terms of confirmed clinical neuropathy, IENFD, NCV or HRQoL. The nerve regeneration rate with glargine was significantly superior than that with exenatide ($p=0.002$ vs. $p=0.06$) [35].

Issar et al. [36] performed a study with five treatment arms: 32 participants treated with exenatide; 31 participants treated with DPP-4is; 27 participants treated with sodium-glucose cotransporter-2 inhibitors (SGLT-2is); 32 healthy controls; and a cohort of ten participants evaluated prospectively for a 3-month period of exenatide treatment. DPP-4is therapy and SGLT-2is therapy had no effect on nerve function; in contrast, subjects treated with exenatide exhibited normal nerve excitability, indicative of impulse conduction. Subjects in the prospective study arm treated with exenatide showed increased S2 accommodation ($p=0.028$), increased superexcitability ($p=0.037$) and diminished subexcitability ($p=0.045$), independent of glycaemic control [36].

## EXPERIMENTAL STUDIES WITH DPP-4IS IN DPN

Experimental studies on DPP-4is in the management of DPN are summarised Table 3.

A recent study by Yamaguchi et al. [37] assessed several DPP-4is (diprotin A, linagliptin, and sitagliptin) and DPP-4 substrates (pituitary adenylate cyclase-activating polypeptide [PACAP], neuropeptide Y [NPY] and stromal cell-derived factor-1a [SDF-1a]) in dorsal root ganglia neurons from C57BL/6J (BL6) mice and 50B11 cells. Diprotin A is a tripeptide only used in the experimental setting [38, 39]. Yamaguchi et al. [37] used neurite outgrowth, a commonly used index, to assess the results, reporting that all three DPP-4is administered showed a significantly elevated mean neurite length, with sitagliptin showing the best result ($p<0.0001$) compared with diprotin A ($p<0.05$) and linagliptin ($p<0.05$). The three DPP-4 substrates administered as potential enhancers of DPP-4is in DPN, namely PACAP, NPY and SDF-1a, are major gut hormones involved in the incretin effect [40]. The results showed that the only substrate significantly increasing neurite growth was PACAP ($p<0.0001$) [37]. There was also an advantageous effect of DPP-4is and PACAP on cell viability: their administration in 50B11 cells (an immortalised cell line of rat dorsal root ganglion neurons) significantly attenuated decreased cell viability under $H_2O_2$ cytotoxic conditions [37].

Jin et al. [41] conducted an experimental study and administered vildagliptin at two different doses, 0.3 mg/kg or 10 mg/kg, on a daily basis to adult male STZ-induced diabetic Sprague–Dawley rats in two of the four experimental groups. In the group treated with vildagliptin at 10 mg/kg/day, nerve fibre loss was significantly limited compared with the other experimental groups ($p<0.05$). Moreover, vildagliptin administration significantly ($p<0.05$) attenuated the decrease in IENFD, evaluated as the number of nerve fibres per millimetre of epidermis in immunohistochemical staining, in a dose-dependent manner. Compared with untreated rats, vildagliptin-treated rats showed a significantly reduced effect at 2000 and 250 Hz ($p<0.05$). These outcomes indicate that sensory

**Table 3** Experimental studies on DPP-4is in DPN

| Study | Animal/experimental model and study design | Major outcomes | Conclusions |
|---|---|---|---|
| Yamaguchi et al. [37] | Primary cultured dorsal root ganglia neurons treated with DPP-4is (diprotin A, linagliptin and sitagliptin) and DPP4 substrates, such as PACAP, NPY and SDF-1a, and 50B11 cells | Mean neurite length was significantly increased with diprotin A ($p < 0.05$), sitagliptin ($p < 0.0001$) and linagliptin ($p < 0.05$)<br>PACAP administartion resulted in a significantly increased neurite growth ($p < 0.0001$), but other DPP-4 substrates showed insignificant changes<br>In 50B11 cells, the decrease in cell viability was suppressed by DPP-4is and PACAP under $H_2O_2$-induced cytotoxic conditions | PACAP upregulation, mediated through DPP-4, inhibition is a promising approach in DPN by promoting neurite elongation in PNS |
| Guo et al. [46] | 5-week old male GK and Wistar rats treated with canagliflozin, teneligliptin or their combination | $V_\beta$ was significantly decreased in GK rats ($p < 0.01$) compared with Wistar rats<br>In GK rats treated with both canagliflozin and teneligliptin, $V_\beta$ was significantly higher ($p < 0.05$); in rats receiving teneligliptin, $V_\beta$ was significantly increased compared with untreated rats ($p < 0.05$)<br>Parasympathetic nerves and IENFD were significantly increased in the treatment arms receiving either canagliflozin or teneligliptin (both $p < 0.05$) and particularly in the combined treatment arm ($p < 0.01$)<br>Parasympathetic nerve density ($r = 0.55$, $p < 0.01$) and IENFD ($r = 0.54$, $p < 0.01$) was associated with $V_\beta$<br>The cut-off value of IENFD for $V_\beta$ estimation was 16.39, yielding 86% sensitivity and 82% specificity | Combined therapy was beneficial; prevention of DPN depends on $V_\beta$ for T2DM without obesity |

**Table 3** continued

| Study | Animal/experimental model and study design | Major outcomes | Conclusions |
|---|---|---|---|
| Sharma et al. [49] | Nicotinamide-STZ induced T2DM Wistar Albino rats treated after day 15 with either metformin, pioglitazone and glimepiride or metformin and sitagliptin or sitagliptin, amitriptyline and sitagliptin | The significantly decreased muscular grip, observed after diabetes induction ($p < 0.001$), was significantly improved for all treatment groups on days 28 and 35 ($p < 0.001$)<br>Paw jumping control, indicative of thermal pain was significantly reduced in all treatment arms on days 21, 28 and 35 ($p < 0.01$), compared with diabetic rats<br>No increase in body weight ($p < 0.01$) and a decrease in blood glucose ($p < 0.001$) were noted in all treatment arms<br>Sciatic nerve samples from rats treated with metformin and sitagliptin showed elevated regeneration capacity, compared with all other study groups | Treatment with sitagliptin or combinations with metformin or amitriptyline increase grip strength and pain sensitivity |
| Bianchi et al. [45] | STZ-induced diabetic rats treated with PKF275-055, a long-lasting vildagliptin analog | PKF275-055 increased $Na^+/K^+$-ATPase activity and also (partially) NCV deficits; no effects on mechanical and thermal sensitivity perception were documented<br>PKF275-055 administration resulted in significantly improved $Na^+/K^+$-ATPase activity and NCV<br>Mechanical sensitivity perception thresholds were significantly improved ($p < 0.01$)<br>A progressive improvement in thermal perception was observed | PKF275-055 emerges as a potential effective treatment of DPN in rodents |

**Table 3** continued

| Study | Animal/experimental model and study design | Major outcomes | Conclusions |
|---|---|---|---|
| Jin et al. [41] | Adult male STZ-induced diabetic Sprague–Dawley rats, allocated in 4 groups each and 2 groups treated with DPP-4is at 2 doses (0.3 mg/kg/day or 10 mg/kg/day) | DPP-4is administration of 10 mg/kg/day resulted in a significant counteraction of nerve fibre loss compared with all other study groups ($p < 0.05$)<br>Both DPP-4is-treated groups showed significantly reduced dose-dependent decrease in IENFD ($p < 0.05$)<br>Compared with untreated DM rats, vildagliptin-treated rats showed significantly reduced effect at 2000 and 250 Hz ($p < 0.05$)<br>No effects on body weight or food intake or blood glucose were documented<br>The highest GLP-1 levels were found in the group treated with the highest DPP-4is dose | Peripheral nerve degeneration is prevented through DPP-4is |

**Table 3** continued

| Study | Animal/experimental model and study design | Major outcomes | Conclusions |
|---|---|---|---|
| Tsuboi et al. [42] | Non-obese T2DM GK and Wistar rats and STZ-induced diabetic mice treated with vildagliptin | Vildagliptin administration resulted in improved glycaemic control, increased insulin and GLP-1 levels, delayed NCV and neuronal atrophy attenuation<br>CGRP-positive cells, previously found decreased in DPN, were increased following vildagliptin administration<br>IENFD was improved following vildagliptin treatment<br>There was restoration of insulin receptor phosphorylation and GLP-1 associated mediators, such as CREB, PKB/AKT and S6RP in the dorsal root ganglion of GK rats<br>In diabetic mice, vildagliptin administration resulted in NCV improvement without any effect on glycaemic control<br>In the dorsal root ganglia of mice, the same effects were observed<br>No effects were observed on insulin receptor β expression | Vildagliptin counteracted insulin resistance in diabetic neurons and improved neuropathy through GLP-1 associated signalling |

*DM* Diabetes mellitus, *DPN* diabetic peripheral neuropathy, *DPP-4* dipeptyl peptidase-4, *DPP-4is* dipeptidyl peptidase 4 inhibitors, *GK* Goto-Kakizaki, *GLP-1* glucagon-like peptide 1, *IENFD* intraepidermal nerve fiber density, *$Na^+/K^+$-ATPase* sodium–potassium adenosine triphosphatase, *NCV* nerve conduction velocity, *NPY* neuropeptide Y, *PACAP* pituitary adenylate cyclase-activating polypeptide, *PKB/AKT* protein kinase B, *PKF275-055* 1-[(2-adamantyl)amino]acetyl-2-cyano-(S)-pyrrolidine, monohydrochloride, *PNS* peripheral nervous system, *S6RP* S6-ribosomal protein, *SDF-1a* stromal cell-derived factor 1a, *STZ* streptozocin, *T2DM* type 2 diabetes mellitus, *$V_\beta$* pancreatic β-cell volume density, *CGRP* calcitonin-gene related peptide, *CREB* cyclic adenosine monophosphate response element binding protein

perception, including pressure, vibration and pain, was significantly improved in the vildagliptin-treated group. The highest measured GLP-1 levels were found in the group treated with the highest vildagliptin dose [41].

The impact of vildagliptin administration on DPN was the objective of a subsequent study by Tsuboi et al. [42]. This basic research study included both non-obese T2DM Goto-Kakizaki (GK) rats and STZ-induced diabetic mice. The results confirmed the beneficial effect of vildagliptin: this agent improved NCV and neuronal atrophy, and also increased IENFD level. Moreover, vildagliptin-based treatment increased the number of calcitonin gene-related peptide (CGRP)-expressing cells [42]. CGRP-expressing neurons promote wound healing and muscle regeneration through inflammation-modulating actions (mainly through inhibition of neutrophil/monocyte/macrophage recruitment and through changes in macrophage polarisation) [43]. An additional vildagliptin-induced effect was the restoration of insulin receptor phosphorylation and GLP-1-associated mediators, such as CREB, PKB/AKT and S6-ribosomal protein (S6RP) in the dorsal root ganglion of GK rats and mice [42]. Alterations in insulin signalling have been reported to promote the development of sensory neuropathy in experimental studies [44].

A long-lasting and orally bioavailable vildagliptin analog termed PKF275-055 (1-[(2-adamantyl)amino]acetyl-2-cyano-(S)-pyrrolidine, monohydrochloride) has also been evaluated in DPN [45]. PKF275-055, when assessed as a prevention agent for DPN, increased $Na^+/K^+$-ATPase activity and also partially reversed NCV deficits without having a major impact on mechanical and thermal sensitivity perception [45]. The administration of PKF275-055 as a therapeutic agent resulted in significantly improved sodium–potassium adenosine triphosphatase ($Na^+/K^+$-ATPase) activity and NCV. Importantly, the thresholds corresponding to mechanical sensitivity perception were significantly improved by almost 50% ($p<0.01$) along with an improvement in thermal perception [45].

In addition to studies focusing on DPP-4i monotherapy in DPN, several combination treatments have been assessed in experimental settings. Guo et al. [46] conducted a study on 5-week-old male GK and Wistar rats and assessed three different therapeutic approaches: one study arm received canagliflozin, a second study arm received teneligliptin and the third study arm received combination therapy with these two DPP-4is. Pancreatic β-cell volume density ($V_\beta$), a novel parameter in this work, was reduced in GK rats ($p<0.01$) compared with Wistar rats, and was significantly increased in rats receiving teneligliptin compared with untreated rats ($p<0.05$). A similar positive effect on $V_\beta$ was noted in GK rats treated with both canagliflozin and teneligliptin ($p<0.05$). Parasympathetic nerves and IENFD were significantly increased in rats receiving either canagliflozin or teneligliptin (both $p<0.05$), particularly in rats in the combined treatment arm ($p<0.01$) [46]. Parasympathetic nerve density ($r=0.55$, $p<0.01$) and IENFD ($r=0.54$, $p<0.01$) were positively associated with $V_\beta$. IENFD was a clinically reliable surrogate marker of $V_\beta$, with a cut-off at 16.39, yielding a sensitivity of 0.86 and specificity of 0.82 [46]. These authors suggested that the effect of canagiflozin in DPN was particularly achieved through major glycaemic regulation, as SGLT-2 is not expressed in the central nervous system [46]. Nevertheless, there is evidence pointing to an effect of SGLT-2is on the nervous system due to their lipid-soluble properties and to their presence in some brain regions [47, 48].

Sharma et al. [49] designed a study encompassing multiple combination treatment and monotherapy regimens with sitagliptin (metformin, pioglitazone and glimepiride or metformin and sitagliptin or sitagliptin, amitriptyline and sitagliptin) using nicotinamide-streptozocin-induced T2DM Wistar Albino rats. Major defects in muscular grip were observed after the induction of diabetes ($p<0.001$). Muscular grip strength was significantly improved in all treatment groups on days 28 and 35 ($p<0.001$). Thermal pain perception was assessed using paw jumping control and was significantly diminished in rats in all treatment arms on days 21, 28 and 35 ($p<0.01$), compared with untreated rats. Finally, increased sciatic regeneration capacity was demonstrated in nerve samples from rats treated with

metformin and sitagliptin, as compared with all other study groups [49].

## CLINICAL STUDIES WITH DPP-4IS IN DPN

Clinical studies on DPP-4is in the management of DPN are summarised in Table 4.

Syngle et al. [50] included 20 T2DM subject in a prospective, open-label, pilot study. In this study, teneligliptin was administered at 20 mg once daily over a period of 3 months. The authors found that therapy reduced the vibration perception threshold (VPT) ($p<0.01$) and that the VPT was associated with C-reactive protein (CRP) ($r=0.69$, $p<0.02$), glycated haemoglobin (HbA1c) ($r=0.48$, $p<0.01$) and heart rate response to standing (HRS) ($r=-0.45$, $p<0.01$). The treatment also improved autonomic neuropathy, as evidenced by sudomotor function ($p<0.01$), HRS ($p<0.01$), heart response to deep breathing (HRD) ($p<0.01$) and blood pressure response to standing (BPS) ($p<0.05$). Teneligliptin improved HRQoL. Significantly reduced glucose, HbA1c, erythrocyte sedimentation rate (ESR) and CRP were seen following teneligliptin treatment [50].

Sitagliptin was evaluated in a 3-month interventional, prospective clinical trial by Barros et al. [51]. These authors observed an improvement in the somatosensory-evoked potentials (SEPs) of the treated participants, as well as a reduction in HbA1c ($p<0.0001$), fasting glucose ($p=0.001$), total cholesterol ($p=0.019$) and alanine transaminase (ALT) ($p=0.022$). Interestingly, the study involved T2DM subjects without clinical manifestations of DPN [51].

A recent 1:5 matched cohort study encompassing data from 2012 to 2021 compared the impact of biguanide- (514 participants) or DPP-4i-based treatment (2570 participants) on DPN [52]. Both agent classes had no significant effect on DPN ($p=0.491$, $p=0.560$ for cumulative incidence after 5 years). A further comparison further substantiated this conclusion, as the hazard ratio for biguanide treatment compared with DPP-4is-based treatment was assessed overall at 0.76 (95% confidence interval [CI] 0.34–1.67) [52].

Kolaczynski et al. [53] included 3015 individuals receiving vildagliptin and 3015 individuals receiving sulfonylurea in a retrospective cohort study on diabetic complications. Vildagliptin administration was linked with a significantly decreased incidence of DPN (odds ratio [OR] 0.71, 95% CI 0.60–0.85, $p=0.0001$), as well as of the composite outcome featuring both retinopathy and DPN (OR 0.70, 95% CI 0.61–0.82, $p<0.0001$) [53].

## DISCUSSION

In this review we discuss current experimental and clinical evidence on the role of GLP-1RAs and DPP-4is in DPN. GLP-1RAs that have been investigated in experimental studies include exenatide [10, 14–17, 19, 21, 22], liraglutide [24] and the twincretin tirzepatide [28]. In these studies, GLP-1R was found to be expressed in the dorsal root ganglion [14], and in two studies GLP-1R expression was not limited to neurons (small and large peptidergic neurons) but was also seen in Schwann cells [20, 22]. In two studies, GLP-1 promoted neurite outgrowth and reduced pro-apoptotic factors (e.g. Bim), induced under NGF deprivation [10, 19]. Interestingly, hippocampal expression of GLP-1R and NGF doubled over a short period of GLP-1RAs-based treatment. In two other studies, NGF expression was restored with liraglutide [24, 27].

In experimental studies, GLP-1RAs achieved its effects by promoting neurite outgrowth, with multiple studies confirming the promotion of neurite outgrowth by GLP-1RAs, their capacity to reassemble of actin and tubulin and their activation of neuronal projections [13, 14, 16, 18–20]. Even increased nerve fibre density in the hind dorsum was seen with exendin-4 treatment, further substantiating the neurotorophic effect of GLP-1RAs [15]. Electrophysiological parameters, such as MNCV and SNCV, have been found to be significantly improved following treatment with GLP-1RAs [14, 22], and myelination markers were also restored [14]. In some studies, a significant improvement was seen in

**Table 4** Clinical studies on DPP-4is in DPN

| Study | Study type | Study population and design | Major outcomes | Conclusions |
|---|---|---|---|---|
| Syngle et al. [50] | Prospective, open-label, pilot study | 20 T2DM subjects with mean age of 56.1 years and mean T2DM duration of 7.75 years treated with teneligliptin 20 mg once daily over 3 months | Significantly high glucose, HbA1c, ESR and CRP were seen<br>HRS and HRD, indicative of cardiovascular autonomic function, were significantly improved (both $p < 0.01$), and HRV improved without reaching significance ($p = 0.12$); HRS was associated with CRP ($r = -0.65, p < 0.01$); CRP was linked with HRV ($r = -0.48, p < 0.01$)<br>Evaluation of the sympathetic function showed significantly improved BPS ($p < 0.05$) and not significantly improved BPH ($p = 0.06$)<br>Sudomotor function was significantly improved ($p < 0.01$); Sudoscan scores were negatively correlated with VPT ($r = -0.49, p = 0.01$), BPS ($r = -0.66, p < 0.001$) and CRP ($r = -0.62, p < 0.01$), and positively correlated with HRV ($r = 0.55, p < 0.01$) and HRS ($r = 0.42, p < 0.01$)<br>Among T2DM subjects, significantly decreased VPT scores were seen ($p < 0.01$); VPT scores were associated with CRP ($r = 0.69, p < 0.02$), HbA1c ($r = 0.48, p < 0.01$) and HRS ($r = -0.45, p < 0.01$)<br>HRQoL was significantly improved ($p < 0.05$) | Teneligliptin improved glycaemic control, alleviated inflammation and ameliorated neuropathy-associated indices in T2DM |
| Nakatani et al. [52] | Cohort study between 2012–2021 with a new-user design and data from Shizuoka Kokuho database | A total of 514 participants with mean age of 68.39 years treated with biguanide, and<br>2570 study participants with mean age of 68.67 years treated with DPP-4i<br>(1:5 matched cohort) | Cumulative incidence of DPN was similar in both study arms ($p = 0.491$, $p = 0.560$ after 5 years)<br>The hazard ratio for biguanide treatment compared with DPP-4is-based treatment was 0.76 (95% CI 0.34–1.67) | Biguanide treatment leads to a similar incidence of DPN and holds similar therapeutic potential as DPP-4is |

**Table 4** continued

| Study | Study type | Study population and design | Major outcomes | Conclusions |
|---|---|---|---|---|
| Kolaczynski et al. [53] | Retrospective cohort study | A total of 3015 out of 16,321 subjects on sulfonylurea treatment with mean age of 63.7 years, mean T2DM duration of 3.2 years and mean treatment duration of 2.5 years, and 3015 out of 4481 subjects on vildagliptin treatment with mean age of 64.6 years, mean T2DM duration of 3.1 years and mean treatment duration of 2.3 years | Vildaglipin treatment was associated with diminished incidence of DPN (OR 0.71, 95% CI 0.60–0.85, $p = 0.0001$) and of the composite outcome including both retinopathy and DPN (OR: 0.70, 95% CI 0.61–0.82, $p < 0.0001$)<br>Incidence rates were not significantly associated with a benefit in a treatment arm<br>In DPN, an incidence rate ratio of 0.99 (95% CI 0.84–1.16, $p = 0.8647$) was observed | Vildagliptin administration was linked with reduced incidence of DPN |
| Barros et al. [51] | Interventional, prospective open clinical trial | 20 T2DM subjects treated with sitagliptin (over 3 months) | Significantly diminished HbA1c ($p < 0.0001$), fasting glucose ($p = 0.001$), total cholesterol ($p = 0.019$) and ALT ($p = 0.022$) were observed<br>At the end of the study, significantly increased GLP-1 activity was observed ($p = 0.0025$)<br>Significant changes in SEPs before and after treatment with sitagliptin were observed | Sitagliptin improves peripheral nerve dysfunction |

*ALT* alanine transaminase, *BPH* blood pressure response to handgrip, *BPS* blood pressure response to standing, *CI* confidence interval, *CRP* C-reactive protein, *DPP-4i(s)* dipeptidyl peptidase 4 inhibitor(s), *ESR* erythrocyte sedimentation rate, *HAQ* health assessment questionnaire, *HbA1c* glycated haemoglobin, *HRD* heart rate response to deep breath, *HRQoL* health-related quality of life, *HRS* heart rate response to standing, *HRV* heart rate response to Valsalva, *GLP-1* glucagon-like receptor agonist, *OR* odds ratio, *SEPs* somatosensory-evoked potentials, *T2DM* type 2 diabetes mellitus, *VPT* vibration perception threshold, *DPN* diabetic peripheral nuropathy

IENFD and a gradual increase in intra-epidermal innervation [13–15, 22]. There has also been evidence of an improvement in neuropathic symptoms: thermal sensory perception was improved with exenatide, whereas the latencies assessed in other exenatide-based studies revealed no effect [21, 22]. A number of studies have shown that GLP-1RAs effectively counteracted oxidative stress and the apoptosis of Schwann cells and/or neurons [15–18, 24].

An interesting action of GLP-1RAs was observed when this agent was co-administered with exendin-4 and LY294002, a PI3K inhibitor, with a further promotion of neurite outgrowth and neuronal survival [20]. GLP-1RAs have also been observed to exhibit immunomodulatory actions, facilitating production of anti-inflammatory cytokines by Schwann cells [24]. Tirzepatide treatment counteracted neuronal apoptosis and facilitated neuronal differentiation [28].

Various experimental studies evaluating DPP-4is have assessed vildagliptin [41, 42], a long-acting vildagliptin analog, termed PKF275-055 [45], diprotin A [37], linagliptin [37], sitagliptin [37, 49], teneligliptin [46] and several combinations of these, such as teneligliptin with canagliflozin [46] or sitagliptin-based combinations (e.g. metformin and sitagliptin, or amitriptyline and sitagliptin) [49]. In one of these studies, vildagliptin emerged as a promising treatment option, effectively reversing NCV delays in the rat model and improving NCV in the mice model [42]. However, mechanical/thermal sensitivity was not improved with vildagliptin [45]. In therapeutic settings, motor thresholds were found to be substantially improved by 50%, whereas a progressive improvement in thermal perception was also noted [45]. Overall sensory function was improved by vildagliptin, showing promising effects in pain, pressure and vibration [41]. Experimental assays resembling clinical tests showed clear benefits of sitagliptin or sitagliptin-based combinations, either in muscular grip or paw jumping control, indicative of thermal pain perception [49].

Evaluations of immunohistochemical parameters further identified novel molecular patterns. Neurite outgrowth was found to be enhanced by several DPP-4is, with sitagliptin yielding the best outcome [37]. Vildagliptin improved IENFD in a dose-dependent manner. Teneligliptin or combined teneligliptin-canagliflozin treatment also resulted in the improvement of this particular index, with the most noteworthy effect specifically found in parasympathetic innervation [41, 46]. Nerve regeneration was observed to be promoted through sitagliptin and metformin treatment [49]. The decrease of CGRP-expressing neurons was counteracted by vildagliptin. The highest GLP-1 levels corresponded to the highest dose of medication administered, while insulin signalling and the action of several GLP-1 downstream mediators (e.g. CREB) were also enhanced [41, 42]. DPP-4 substrates and their introduction into clinical practice may also pave the way for a more efficient therapeutic approach in treating subjects with DPN, as PACAP administration further strengthened neurite outgrowth [37].

Many clinical studies shared common methodology in terms of participant selection. Clinical diagnosis was established according to the criteria of the American Diabetic Association [30, 50], the International Classification of Diseases (ICD-10) [43], the Michigan Diabetes Neuropathy Scale [35], the TNS [33, 34, 36] or mTCNS [33]. Most studies also adjusted for potential alternative diagnoses by excluding subjects suffering from vitamin $B_{12}$ deficiency [33], those who had been administered neurotoxic agents (e.g. immunotherapy, chemotherapy) [33, 34], those who consumed alcohol [52] and neuropathy attributed to other reasons [32, 34].

The number of studies on GLP-1RAs is limited although some data are available from studies ranging in designs (observational [30, 32, 34], comparative [36] and a 18-month proof-of-concept open-label randomised study [35]). Semaglutide [32], dulaglutide [32] and exenatide [34, 35] have been used so far. In these studies, treatment with GLP-1RAs resulted in major improvement in CSA [32]. Improvement in $Na^+$/$K^+$-ATPase pump function and $Na^+$ conduction, indicative of a more optimal neuronal function, was found in experiments carried out in clinical settings, and also confirmed in experimental works [16, 34]. Despite substantially increased sural amplitude, NCV was not affected [32, 34]. Excitability assays have also been carried out, with the results indicating that only exenatide

**Table 5** Emerging mechanisms of incretin action in DPN in experimental studies.

| Agent class | Potential mechanisms of action | Outcomes in experimental studies | References |
|---|---|---|---|
| DPP-4is | Increased GLP-1 levels<br>Increase in CGRP expressing neurons<br>Restoration of insulin signalling<br>Improved activity of $Na^+$/$K^+$-ATPase<br>Enhanced action with PACAP co-administration | Improvement of IENFD<br>Increased neurite outgrowth<br>Increased cell viability under cytotoxic conditions<br>Increased $V_\beta$<br>Increased NCV<br>Improved mechanical sensitivity and thermal pain perception<br>Increased nerve regeneration capacity<br>Increased muscular grip strength | [37, 41, 42, 45, 46, 49] |
| GLP-1RAs | Attenuation of oxidative stress<br>Alterations in actin and tubulin<br>Increased cell membrane surface, increased $Na^+$ conduction and increased $Ca^{2+}$ currents<br>Induction of Epac/AKT pathway<br>NGF-like action<br>RhoA suppression<br>NF-κB suppression<br>Enhanced action with PI3K inhibition<br>Increased pERK1/2 | Improvement of sensory and motor NCV<br>Increased neurite outgrowth and neurite number<br>Alleviation of neuropathic symptoms (e.g. hypoalgesia)<br>Increased IENFD<br>Increased myelinated fibre size and decreased axon/fibre area ratio<br>Increased nerve fibre density in the hind dorsum<br>Decrease of Schwann cell apoptosis<br>Increase of cAMP levels<br>Induction of mature neuronal phenotype<br>Improved mitchondrial function<br>Induction of cell-proliferation and neurite formation<br>Inhibition of pro-apoptotic proteins and of ROS defence markers levels<br>Increased expression of anti-apoptotic markers<br>Improved pain perception<br>Improved myelination | [10, 13–22, 24, 27] |
| GLP-1/GIP receptor agonists | Increased neurotrophic factors<br>Activation of pAKT/CREB/BDNF pathway<br>Epigenetic modulation of neuronal growth, apoptosis and differentiation | Increase in GLUT3 and GLUT4<br>Restoration of insulin signalling<br>Increased neuroprotection capacity | [28] |

*AKT* Protein kinase B, *BDNF* brain derive neurotrophic factor, *cAMP* cyclic adenosine monophosphate, *CGRP* calcitonin gene-related peptide, *CREB* cyclic adenosine monophosphate response element-binding protein, *DPP-4is* dipeptidyl peptidase 4 inhibitors, *GIP* glucose-dependent insulinotropic peptide, *GLP-1* glucagon-like peptide 1, *GLUT* glucose transporter, *IENFD* intra-epidermal nerve fibre density, *$Na^+/K^+$-ATPase* sodium–potassium adenosine triphosphatase, *NCV* nerve conduction velocity, NF-κB, nuclear factor kappa-light-chain-enhancer of activated B cells, *NGF* nerve growth factor, *PACAP* pituitary adenylate cyclase-activating polypeptide, *pERK1/2* proline extensin-like receptor kinase 1/2, *PI3K* phosphoinositide 3-kinase, *RhoA* Ras homolog family member A, *ROS* reactive oxygen species, $V_\beta$ pancreatic β-cell volume density, *DPN* diabetic peripheral neuropathy

could contribute effectively to optimal neuronal function, compared to both DPP-4is and SGLT-2is [36]. In some studies, assessment using the TNS yielded contradictory data with improvement in two studies [32, 34] and a lack of improvement, at least in motor modalities, in a third study [36]. Again, exenatide was inferior to insulin glargine in one study but effective in other studies [34, 35]. In one study, semaglutide and dulaglutide were compared, with the authors reporting a non-significant trend in favour of dulaglutide with respect to tibial CSA measurements [32]. The effect of GLP-1RAs on autonomic nerve function has not been fully elucidated, and only limited data exist. On the one hand, a beneficial effect on autonomic neuropathy has been assessed [50]; on the other hand, the known inhibitory effect of GLP-1RAs on gastrointestinal motility has not been evaluated and the exact effect needs to be clarified [54].

Clinical data on DPP-4is in DPN are also sparse. Overall, data from two studies with a prospective design (a prospective, open-label, pilot study [50] and an interventional, prospective and open clinical trial [51]), one matched cohort study [52] and one retrospective cohort study [53] are available. These studies used sitagliptin [51], vildagliptin [53] or teneligliptin [50]. In one study, a substantial decrease in VPT was observed in teneligliptin-treated individuals, showing that DPP-4is-based treatment alleviated large myelinated fibre dysfunction [50]. In another study, the improvement in SEPs, corresponding to restored peripheral nerve function under sitagliptin treatment, was also considerable [51]. Of note, one study strengthened the potential role of DPP-4is in cardiac autonomic neuropathy, yielding promising outcomes in multiple associated indices [50]. There is increasing evidence pointing to the need for further studies to elucidate the role of DPP-4is in DPN: beyond significant improvement in HRQoL, vildagliptin was found to significantly reduce the incidence of DPN restrospectively and prevent the occurrence of combined outcome including DPN and retinopathy [50, 53]. Perhaps, the reason for the limited clinical evidence of DPP-4is lies in their lack of benefit, as compared with other medications, such as biguanides [52].

The effect of incretins on DPN is very promising, both in clinical and experimental settings. Incretins essentially improve neuronal function, counteract inflammation and oxidative stress and imitate or promote the actions of several neurotrophic factors. An overview of the multiple mechanisms mediating the benefit of DPP4-is and GLP-1RAs in DPN can be found in Table 5. Compared with other antidiabetic agents, the potential of incretins is more promising for different reasons. Metformin has been associated with vitamin $B_{12}$ deficiency, which may per se lead to neuropathy. Consequently, great caution is needed [55]. Conversely, the benefit documented for canagliflozin has been mainly attributed to the restoration of normoglycaemia and not to any effects specific to nervous system, as the expression of these cotransporters in the nerve system is ambiguous [46–48].

## STRENGTHS AND LIMITATIONS OF THIS REVIEW

The strength of this review is inclusion of both experimental and clinical data. Also, the clinical outcomes related to DPN are provided in a time-dependent way, showing the effects of DPP-4is and GLP-1RAs in subjects with DPN. Nevertheless, there are certain limitations. First, participant series in clinical studies were small. Moreover, most studies were not interventional but relied on retrospective data. Accordingly, they could not fully document any benefit for DPP-4is and GLP-1RAs, but only compare their effects with other antidiabetic agents. Thirdly, design, research questions and study endpoints were heterogeneous. Indeed, even contradictory results have been reported.

## CONCLUSIONS

The role of GLP-1RAs and DPP-4is in DPP has not been sufficiently studied so far. Experimental evidence suggests that both agent classes not only enhance neurite outgrowth, promote increased innervation and restore motor and sensory electrophysiological parameters, but

they also counteract the detrimental effects of oxidative stress and inflammation inducing apoptosis. Clinical data are still sparse, but promising, pointing to a beneficial effect on electrophysiological parameters, neuronal function and HRQoL. Nevertheless, the limited number of participants in the clinical trials and the relative scarcity of evidence pose considerable obstacles in terms of the clinical utility of antidiabetic agents to improve DPN-associated outcomes. Therefore, further studies are required, which will determine the exact effect of both drug categories on their utility in DPN and whether their actions are dependent on any associated parameters of the condition, such as severity and/or symptoms. Finally, studies should shed light on the question of whether DPP-4is and/or GLP-1RAs hold potential as preventive agents for DPN, given that they are widely prescribed among T2DM subjects.

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## REFERENCES

1. Hicks CW, Selvin E. Epidemiology of peripheral neuropathy and lower extremity disease in diabetes. Curr Diab Rep. 2019;19(10):86.

2. American Diabetes Association Professional Practice Committee. 12. Retinopathy, neuropathy, and foot care: standards of care in diabetes-2025. Diabetes Care. 2025;48(Supplement_1):S252–65.

3. Savelieff MG, Elafros MA, Viswanathan V, Jensen TS, Bennett DL, Feldman EL. The global and regional burden of diabetic peripheral neuropathy. Nat Rev Neurol. 2024;1:17–31. https://doi.org/10.1038/s41582-024-01041-y.

4. Goldney J, Sargeant JA, Davies MJ. Incretins and microvascular complications of diabetes: neuropathy, nephropathy, retinopathy and microangiopathy. Diabetologia. 2023;66(10):1832–45.

5. Hemmingsen B, Sonne DP, Metzendorf MI, Richter B. Dipeptidyl-peptidase (DPP)-4 inhibitors and glucagon-like peptide (GLP)-1 analogues for prevention or delay of type 2 diabetes mellitus and its associated complications in people at increased risk for the development of type 2 diabetes mellitus. Cochrane Database Syst Rev. 2017;5(5):CD012204.

6. García-Casares N, González-González G, de la Cruz-Cosme C, et al. Effects of GLP-1 receptor agonists on neurological complications of diabetes. Rev Endocr Metab Disord. 2023;24(4):655–72.

7. Nauck MA, Meier JJ. The incretin effect in healthy individuals and those with type 2 diabetes: physiology, pathophysiology, and response to therapeutic interventions. Lancet Diabetes Endocrinol. 2016;4(6):525–36.

8. Baik SH, McDonald CJ. Independent effects of 15 commonly prescribed drugs on all-cause mortality among US elderly patients with type 2 diabetes mellitus. BMJ Open Diabetes Res Care. 2020;8(1):e000940.

9. Gouveri E, Popovic DS, Papanas N. Potential new therapeutic implications of semaglutide: new colours of the rainbow? Diabetes Ther. 2024;15(1):13–8.

10. Biswas SC, Buteau J, Greene LA. Glucagon-like peptide-1 (GLP-1) diminishes neuronal degeneration and death caused by NGF deprivation by suppressing Bim induction. Neurochem Res. 2008;33(9):1845–51.

11. Exenatide: AC 2993, AC002993, AC2993A, exendin 4, LY2148568. Drugs R D. 2004;5(1):35–40.

12. Yap MKK, Misuan N. Exendin-4 from Heloderma suspectum venom: from discovery to its latest application as type II diabetes combatant. Basic Clin Pharmacol Toxicol. 2019;124(5):513–27.

13. Kan M, Guo G, Singh B, Singh V, Zochodne DW. Glucagon-like peptide 1, insulin, sensory neurons, and diabetic neuropathy. J Neuropathol Exp Neurol. 2012;71(6):494–510.

14. Himeno T, Kamiya H, Naruse K, et al. Beneficial effects of exendin-4 on experimental polyneuropathy in diabetic mice. Diabetes. 2011;60(9):2397–406.

15. Liu WJ, Jin HY, Lee KA, Xie SH, Baek HS, Park TS. Neuroprotective effect of the glucagon-like peptide-1 receptor agonist, synthetic exendin-4, in streptozotocin-induced diabetic rats. Br J Pharmacol. 2011;164(5):1410–20.

16. Luciani P, Deledda C, Benvenuti S, et al. Differentiating effects of the glucagon-like peptide-1 analogue exendin-4 in a human neuronal cell model. Cell Mol Life Sci. 2010;67(21):3711–23.

17. Pandey S, Mangmool S, Madreiter-Sokolowski CT, Wichaiyo S, Luangmonkong T, Parichatikanond W. Exendin-4 protects against high glucose-induced mitochondrial dysfunction and oxidative stress in SH-SY5Y neuroblastoma cells through GLP-1 receptor/Epac/Akt signalling. Eur J Pharmacol. 2023;954: 175896.

18. Mohiuddin MS, Himeno T, Inoue R, et al. Glucagon-like peptide-1 receptor agonist protects dorsal root ganglion neurons against oxidative insult. J Diabetes Res. 2019;2019:9426014.

19. Perry T, Lahiri DK, Chen D, et al. A novel neurotrophic property of glucagon-like peptide 1: a promoter of nerve growth factor-mediated differentiation in PC12 cells. J Pharmacol Exp Ther. 2002;300(3):958–66.

20. Tsukamoto M, Niimi N, Sango K, Takaku S, Kanazawa Y, Utsunomiya K. Neurotrophic and neuroprotective properties of exendin-4 in adult rat dorsal

root ganglion neurons: involvement of insulin and RhoA. Histochem Cell Biol. 2015;144(3):249–59.

21. Komsuoglu Celikyurt I, Mutlu O, Ulak G, et al. Exenatide treatment exerts anxiolytic- and antidepressant-like effects and reverses neuropathy in a mouse model of type-2 diabetes. Med Sci Monit Basic Res. 2014;20:112–7.

22. Jolivalt CG, Fineman M, Deacon CF, Carr RD, Calcutt NA. GLP-1 signals via ERK in peripheral nerve and prevents nerve dysfunction in diabetic mice. Diabetes Obes Metab. 2011;13(11):990–1000.

23. Gasbjerg LS, Bari EJ, Christensen M, Knop FK. Exendin(9–39)NH2: Recommendations for clinical use based on a systematic literature review. Diabetes Obes Metab. 2021;23(11):2419–36.

24. Kornelius E, Tsou SH, Chang CC, et al. Liraglutide attenuates glucolipotoxicity-induced rsc96 schwann cells' inflammation and dysfunction. Biomolecules. 2022;12(10):1338.

25. Tomlinson DR, Fernyhough P, Diemel LT. Neurotrophins and peripheral neuropathy. Phil Trans R Soc Lond B. 1996;351(1338):455–62.

26. Gaur A, Varatharajan S, Balan Y, et al. Brain-derived neurotrophic factor (BDNF) and other neurotrophic factors in type 2 diabetes mellitus and their association with neuropathy. Ir J Med Sci. 2024;193(5):2287–92.

27. Gumuslu E, Cine N, Ertan M, Mutlu O, Komsuoglu Celikyurt I, Ulak G. Exenatide upregulates gene expression of glucagon-like peptide-1 receptor and nerve growth factor in streptozotocin/nicotinamide-induced diabetic mice. Fundam Clin Pharmacol. 2018;32(2):174–80.

28. Fontanella RA, Ghosh P, Pesapane A, et al. Tirzepatide prevents neurodegeneration through multiple molecular pathways. J Transl Med. 2024;22(1):114.

29. Tan TM, Khoo B. Tirzepatide and the new era of twincretins for diabetes. Lancet. 2021;398(10295):95–7.

30. Senyigit A, Tabak O, Orhanoglu T, et al. Glucagon-like peptide-1 levels and dipeptidyl peptidase-4 activity in type 2 diabetes. Clin Invest Med. 2017;40(5):E188–99.

31. Hörbelt T, Knebel B, Fahlbusch P, et al. The adipokine sFRP4 induces insulin resistance and lipogenesis in the liver. Biochim Biophys Acta Mol Basis Dis. 2019;1865(10):2671–84.

32. Dhanapalaratnam R, Issar T, Lee ATK, et al. Glucagon-like peptide-1 receptor agonists reverse nerve morphological abnormalities in diabetic peripheral neuropathy. Diabetologia. 2024;67(3):561–6.

33. Dhanapalaratnam R, Issar T, Poynten AM, Milner KL, Kwai NCG, Krishnan AV. Diagnostic accuracy of nerve ultrasonography for the detection of peripheral neuropathy in type 2 diabetes. Eur J Neurol. 2022;29(12):3571–9.

34. Dhanapalaratnam R, Issar T, Poynten AM, Milner KL, Kwai NC, Krishnan AV. Impact of glucagon-like peptide-1 receptor agonists on axonal function in diabetic peripheral neuropathy. J Neurophysiol. 2025;133(1):14–21.

35. Jaiswal M, Martin CL, Brown MB, et al. Effects of exenatide on measures of diabetic neuropathy in subjects with type 2 diabetes: results from an 18-month proof-of-concept open-label randomized study. J Diabetes Complications. 2015;29(8):1287–94.

36. Issar T, Kwai NCG, Poynten AM, Arnold R, Milner KL, Krishnan AV. Effect of exenatide on peripheral nerve excitability in type 2 diabetes. Clin Neurophysiol. 2021;132(10):2532–9.

37. Yamaguchi M, Noda-Asano S, Inoue R, et al. Dipeptidyl peptidase (DPP)-4 inhibitors and pituitary adenylate cyclase-activating polypeptide, a DPP-4 substrate, extend neurite outgrowth of mouse dorsal root ganglia neurons: a promising approach in diabetic polyneuropathy treatment. Int J Mol Sci. 2024;25:8881.

38. Rahfeld J, Schierhorn M, Hartrodt B, Neubert K, Heins J. Are diprotin A (Ile-Pro-Ile) and diprotin B (Val-Pro-Leu) inhibitors or substrates of dipeptidyl peptidase IV? Biochim Biophys Acta. 1991;1076(2):314–6.

39. Alponti RF, Frezzatti R, Barone JM, Alegre Vde S, Silveira PF. Dipeptidyl peptidase IV in the hypothalamus and hippocampus of monosodium glutamate obese and food-deprived rats. Metabolism. 2011;60(2):234–42.

40. Meier JJ, Nauck MA. Incretin-based therapies: where will we be 50 years from now? Diabetologia. 2015;58(8):1745–50.

41. Jin HY, Liu WJ, Park JH, Baek HS, Park TS. Effect of dipeptidyl peptidase-IV (DPP-IV) inhibitor (vildagliptin) on peripheral nerves in streptozotocin-induced diabetic rats. Arch Med Res. 2009;40:536–44.

42. Tsuboi K, Mizukami H, Inaba W, Baba M, Yagihashi S. The dipeptidyl peptidase IV inhibitor vildagliptin suppresses development of neuropathy in diabetic rodents: effects on peripheral sensory nerve

function, structure and molecular changes. J Neurochem. 2016;136:859–70.

43. Lu YZ, Nayer B, Singh SK, et al. CGRP sensory neurons promote tissue healing via neutrophils and macrophages. Nature. 2024;628(8008):604–11.

44. Hackett AR, Strickland A, Milbrandt J. Disrupting insulin signalling in Schwann cells impairs myelination and induces a sensory neuropathy. Glia. 2020;68(5):963–78.

45. Bianchi R, Cervellini I, Porretta-Serapiglia C, et al. Beneficial effects of PKF275-055, a novel, selective, orally bioavailable, long-acting dipeptidyl peptidase IV inhibitor in streptozotocin-induced diabetic peripheral neuropathy. J Pharmacol Exp Ther. 2012;340:64–72.

46. Guo D, Mizukami H, Osonoi S, et al. Beneficial effects of combination therapy of canagliflozin and teneligliptin on diabetic polyneuropathy and β-cell volume density in spontaneously type 2 diabetic Goto-Kakizaki rats. Metabolism. 2020;107: 154232.

47. Wright EM, Loo DD, Hirayama BA. Biology of human sodium glucose transporters. Physiol Rev. 2011;91(2):733–94.

48. Poppe R, Karbach U, Gambaryan S, et al. Expression of the Na+-D-glucose cotransporter SGLT1 in neurons. J Neurochem. 1997;69(1):84–94.

49. Sharma AK, Sharma A, Kumari R, et al. Sitagliptin, sitagliptin and metformin, or sitagliptin and amitriptyline attenuate streptozotocin-nicotinamide induced diabetic neuropathy in rats. J Biomed Res. 2012;26:200–10.

50. Syngle A, Chahal S, Vohra K. Efficacy and tolerability of DPP4 inhibitor, teneligliptin, on autonomic and peripheral neuropathy in type 2 diabetes: an open label, pilot study. Neurol Sci. 2021;42(4):1429–36.

51. Barros JI, Fechine FV, Montenegro Júnior RM, et al. Effect of treatment with sitagliptin on somatosensory-evoked potentials and metabolic control in patients with type 2 diabetes mellitus. Arq Bras Endocrinol Metabol. 2014;58(4):369–76.

52. Nakatani E, Ohno H, Satoh T, et al. Comparing the effects of biguanides and dipeptidyl peptidase-4 inhibitors on cardio-cerebrovascular outcomes, nephropathy, retinopathy, neuropathy, and treatment costs in diabetic patients. PLoS ONE. 2024;19(8): e0308734.

53. Kolaczynski WM, Hankins M, Ong SH, Richter H, Clemens A, Toussi M. Microvascular outcomes in patients with type 2 diabetes treated with vildagliptin vs. sulfonylurea: a retrospective study using German electronic medical records. Diabetes Ther. 2016;7(3):483–496.

54. Hellström PM. Glucagon-like peptide-1 gastrointestinal regulatory role in metabolism and motility. Vitam Horm. 2010;84:319–29.

55. Bell DSH. Metformin-induced vitamin B12 deficiency can cause or worsen distal symmetrical, autonomic and cardiac neuropathy in the patient with diabetes. Diabetes Obes Metab. 2022;24(8):1423–8.