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Mechanism of the neuroprotective effect of GLP-1 in a rat model of Parkinson's with pre-existing diabetes

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ABSTRACT

Several studies have suggested the association between neurodegenerative diseases and diabetes mellitus (DM), DM causes cognitive impairment with age, but its effect is not well known in Parkinson's disease (PD). As a member of the incretin family, Glucagon-like peptide-1 (GLP-1) has glycemic regulation functions. It also exerts many additional effects on different tissues through its receptor's widespread expression.

Objective: our aim is to investigate the effect of pre-existing diabetes on the severity of PD in male albino rats, and to find out whether GLP-1 could improve PD symptoms in diabetic animals in addition to its hypoglycemic effect, and how it could do that.

Methods: 75 adult male albino rats were equally divided into: Control, Parkinson's, Diabetic Parkinson's, Diabetic Parkinson's + low dose exenatide (GLP-1 receptor agonist), Diabetic Parkinson's + high dose exenatide group. Blood glucose and insulin, striatal dopamine, some striatal oxidative stress and inflammatory markers, and the catalepsy score were measured.

Results: Pre-existing of diabetes before initiation of PD raises the severity of PD shown by the more significant increase in catalepsy score, and the more significant decrease in striatal dopamine level. GLP-1 effects extend beyond their hypoglycemic effects only since it has a direct anti-oxidant, and anti-inflammatory neuronal effect with increasing the striatal dopamine and improving the catalepsy score in a dose dependent manner.

Conclusions: Diabetes increases the severity of impairment in PD, and GLP-1 improve it through its direct neuronal effect in addition to its indirect effect through producing hypoglycemia.

1. Introduction

Parkinson's disease (PD) is one of the most common chronic neurodegenerative disorder following Alzheimer's disease which affects more than 1% of the world's elderly (Pringsheim et al., 2014). It is a progressive, disabling motor disorder, its symptoms (bradykinesia, rigidity, resting tremor, and postural instability) are mostly due to the reduction in substantia nigra dopaminergic activity in the midbrain (Sethi, 2008).

In spite of the availability of multiple effective symptomatic drugs, there is no treatment for PD and all attempts have been ineffective to slow down the loss of neuronal cells. This may be due to the fact that substantia nigra pars compacta (SNc) homeostasis is susceptible to various environmental, cellular, and genetic factors which individually or concurrently lead to death of the cell overtime (Lang and Espay, 2018).

The PD pathogenesis research has identified a number of potential contributing factors in the earlier 30 years, indicating that it has multifactorial origin. Beside numeral genetic mutations, metabolic and dietary factors appear to be complicated. Essential stages in this neurodegenerative pathway have been identified demonstrating the relationship between PD pathogenesis and the mechanisms underlying the production of insulin resistance (Cereda et al., 2011). While other studies have indicated that PD's pathogenic mechanisms also include oxidative stress (Manoharan et al., 2016), inflammation (Taylor et al., 2013), and apoptosis (Venderova and Park, 2012).

Incretins are a group of metabolic hormones promoting the reduction of blood glucose levels (Amori et al., 2007). There are two major candidate molecules which carry out incretin criteria; the Gastric inhibitory peptide (GIP) and the intestinal peptide Glucagon-like peptide-1 (GLP-1) (Drucker and Nauck, 2006).

GLP-1 is an intestinal gut hormone that is secreted in a response to food intake and potentiates the glucose-dependent insulin secretion from the pancreatic beta-cells. Moreover, GLP-1 suppresses glucagon

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secretion from alpha-cells, resulting in glucose-dependent reduction in the production of hepatic glucose (Holst et al., 2011). As well as enhancing glycemic control, GLP-1 receptor agonists also develop weight loss (Gerich, 2013). However, the collection of data from preclinical and clinical studies indicates that the effects of GLP-1 receptor agonists go beyond weight loss and glycemic control alone (Seufert and Gallwitz, 2014).

GLP-1 receptors are located widely in many tissues outside the pancreas, such as the gastrointestinal system, kidneys, central nervous system and cardiovascular system, (Pyke et al., 2014). It is therefore reasonable to consider that many physiological effects are mediated by GLP-1 receptor agonists, regardless of their key actions to enhance glycemic control and promote loss of body weight.

In the central nervous system (CNS), GLP-1 was associated with the control of food intake (Barrera et al., 2011), the role of the hypothalamic-pituitary-adrenal axis (Ghosal et al., 2013), the activation of sympathetic nervous system (SNS) (Yamamoto et al., 2002), and visceral dysfunction (Kinzig et al., 2002). Nonetheless, there are still unknown additional effects of the central stimulation of GLP-1. Several clinical trials indicated that drugs used in diabetes treatment had beneficial effects on neurodegenerative processes and on clinical outcome, regarding memory and cognition, and could potentially be translated into new therapies against PD and related conditions (Green et al., 2019), and some of them reported the protective effects of the GLP-1 mimetic exendin-4 in patients with Parkinson's disease (Athauda et al. 2017, 2019; Athauda and Foltynie, 2018). However, the number of these clinical studies is low and further clinical trials are needed.

The aim of this work is to investigate the effect of preexisting diabetes induction on the severity of experimentally induced Parkinson's disease in male rats, and to determine whether GLP-1 can reverse symptoms of Parkinson's disease in the diabetic animals beside controlling hyperglycemia, and work out how it might do this.

2. Materials and methods

2.1. Ethics statement and animals

Research procedure was approved by Faculty of Medicine Research Ethics Committee (FMREC), Minia University, and carried out in compliance with the National Institutes of Guide for the Care and Use of Laboratory Animals (Manual and Manual, 2010). All precautions taken to diminish the number and suffering of animals used in the experiment.

Seventy five (specific-pathogen-free) adult male albino Sprague–Dawley rats that weighed about 150–200 g were used. The rats received from the Faculty of Medicine, Minia University animal House. All rats housed with normal light/dark cycles at room temperature, water and food allowed ad-libitum. Rats randomly divided into five equal groups as follows:

I. Control group: vehicle-injected group receiving normal saline daily for 28 successive days by intraperitoneal injection.
II. Parkinson's group: rats received 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) that dissolved in normal saline by intraperitoneal injection in a dosage of 30 mg/kg body weight at 24 h intervals for 28 consecutive days (Lee et al., 2011).
III. Diabetic Parkinson's group: diabetic induction by injection of 50 mg/kg streptozotocin (STZ) single intraperitoneal injection (Szkudelski, 2001) four days before the start of MPTP-induced Parkinson's.
IV. Diabetic Parkinson's + low dose exenatide (GLP-1 receptor agonist) group: STZ diabetic induction four days before the start of MPTP Parkinson's induction + treating the rats with 1.0 μg/kg single subcutaneous exenatide daily dose on the 4th day of STZ treatment for 28 consecutive days (Elbassuoni, 2014).
V. Diabetic Parkinson's + high dose exenatide group: STZ diabetic induction four days before the start of MPTP Parkinson's induction + treating the rats with 5.0 μg/kg single subcutaneous exenatide daily dose on the 4th day of STZ treatment for 28 consecutive days (Elbassuoni, 2014).

2.2. Induction of diabetes

Diabetes induction by injection of STZ (50 mg/kg). It was freshly dissolved in sodium citrate solution (pH 4.5) and intraperitoneally injected in a single dose (Szkudelski, 2001). Tail vein blood samples taken for blood glucose level analysis after four days of STZ injection. Rats with a fasting blood glucose level above 180 mg/DL (10 mmol/l) are considered to be diabetic.

2.3. Behavioral tests

PD development was observed after 28 days from induction with MPTP, by the occurrence of tremors and the observation of rigidity and bradykinesia in rats that further quantified by ‘‘Catalepsy test‘‘. The first part of this test was the grid test in which the rat was hung on a vertical grid by its paws (the grid was 44 cm high and 25.5 cm wide with a space of one cm in between each wire), time reported for each rat to move its paws or any other sort of the first movement. The second part of the test was the bar test where the rat placed on a bar (9 cm above and parallel from the base) with both fore paws, the time of removal of the paw recorded (Alam and Schmidt, 2004). For all the rats included in the study, these two tests were performed. Several studies have indicated that these tests are sensitive methods for evaluating motor dysfunctions in the MPTP animal model of Parkinson's disease (Alam and Schmidt, 2002; Abdin and Hamouda, 2008; Kim et al., 2010).

2.4. Biochemical analyses of the blood and brain tissue homogenate

Four rats died from the Diabetic Parkinson's group and this could be due to the severe hyperglycemia, since this group have not received any hypoglycemic drugs for the 28 days length of the study. Only 10 rats were included from each group in the statistical analysis to standard the number of animals in all groups.

At the end of the work and after conducting the behavioral tests, the rats sacrificed; blood samples were immediately collected in 10-ml Eppendorf tubes, left to clot, then transferred into centrifuge tubes and centrifuged for 20 min at 3000 rpm; serum samples separated in 2-ml Eppendorf tubes to be used immediately as fresh samples (preferred) or to be stored on − 20 °C until used. Samples of the serum used to determine serum glucose using colorimetric assay kit from MyBioSource, USA, insulin level using ELISA kit from MyBioSource, USA.

The brains were immediately removed and washed with ice-cold saline, each brain's hemisphere striata isolated, weighed, and homogenized with a homogenizer. Homogenization carried out in phosphate-buffered saline (pH = 7.4). After 10 min, the homogeneous centrifuged. The supernatants kept at – 80 °C until the analysis of: dopamine using Rat Dopamine (DA) ELISA Kit from CUSABIO, USA; malondialdehyde (MDA) using colorimetric assay kit from MyBioSource, USA; Superoxide Dismutase Activity using colorimetric assay kit (ab65354) from Abcam, USA; Catalase Activity using colorimetric assay kit (ab83464) from Abcam, USA; Reduced Glutathione (GSH) using colorimetric assay kit (ab235670) from Abcam, USA; tumor necrosis factor-α (TNF-α) by ELISA kit from MyBioSource, USA; Interleukin 1 beta (IL-1β) using ELISA Kit (ab100768) from Abcam, USA.

Streptozotocin, MPTP, exenatide, and other chemicals obtained from Sigma–Aldrich Chemical Co.

2.5. Statistical analysis

Values expressed as mean ± SEM, statistically evaluated using SPSS, version 17 (SPSS Inc., Chicago, IL, USA). For all analyses, one-

Table 1
Striatal dopamine level in the different studied groups.

Parameter	Control Group	Parkinson's Group	Diabetic Parkinson's Group	Diabetic Parkinson's + Low Dose Exenatide Group	Diabetic Parkinson's + High Dose Exenatide Group
Dopamine (ng/ml)	7.3 ± 0.7	3.5 ± 0.5[a]	1.2 ± 0.2[a,b]	2.8 ± 0.4[a,c]	5.4 ± 0.5[a,b,c,d]

Values expressed as mean ± SEM. $n = 10$, $P < 0.05$ was considered statistically significant.
[a] Significant from control group.
[b] Significant from Parkinson's group.
[c] Significant from Diabetic Parkinson's group.
[d] Significant from Diabetic Parkinson's + Low Dose Exenatide group.

way analysis of variance (ANOVA) followed by multiple comparisons test were applied. P value less than 0.05 was considered statistically significant.

3. Results

3.1. Assessment of motor function

Parkinson's group exhibited a significant increase in catalepsy score of either grid test or bar test compared to control group. Diabetic Parkinson's group receiving no treatment showed more significant increase in catalepsy score of either grid test or bar test compared to Parkinson's group. Treatment of Diabetic Parkinson's group with exenatide showed a significant reduction in the catalepsy score of grid test and bar test in a dose-dependent way as compared to Diabetic Parkinson's group receiving no treatment (Table 1).

3.2. Biochemical analysis

3.2.1. Striatal dopamine content

Parkinson's group showed a significant decrease in striatal dopamine levels compared to control group. Diabetic Parkinson's group receiving no treatment showed more decrease in striatal dopamine level compared to Parkinson's group. Treatment of Diabetic Parkinson's group with exenatide significantly improved the striatal dopamine level in a dose-dependent way as compared to Diabetic Parkinson's group receiving no treatment (Table 2).

3.2.2. Serum glucose and insulin

Parkinson's group showed no significant change in serum glucose and insulin levels compared to control group. Diabetic Parkinson's group receiving no treatment showed a significant increase in serum glucose level and significant decrease in serum insulin level compared to Parkinson's group. Treatment of Diabetic Parkinson's group with exenatide significantly decrease serum glucose level, and significantly increase serum insulin level in a dose-dependent way when compared to Diabetic Parkinson's group receiving no treatment (Table 3).

3.2.3. Striatal oxidative stress biomarkers

Measuring MDA level, as a stable product of lipid peroxidation, is a reliable tool to assess the extent of oxidative damage at the cellular level. As compared to the control group, there was a significant increase in striatal MDA level in the Parkinson's group, with more increase in its level in the Diabetic Parkinson's group. Treatment of Diabetic Parkinson's group with exenatide significantly decreased the striatal MDA level in a dose-dependent way as compared to Diabetic Parkinson's group receiving no treatment.

The Parkinson's group exhibited a significant decrease in the activities of the antioxidant enzymes SOD, CAT and GSH in the striata compared to the control group, with more decrease in their activities in the Diabetic Parkinson's group. Treatment of Diabetic Parkinson's group with exenatide significantly increased their activities in the striata in a dose-dependent way as compared to Diabetic Parkinson's group receiving no treatment (Fig. 1).

3.2.4. Striatal inflammatory biomarkers

In the present study, striatal TNF-α and IL-1β levels were significantly increased in the Parkinson's group compared to the control group, with a more significant increase in the Diabetic Parkinson's group as compared to Parkinson's group. Treatment with exenatide significantly decreased TNF-α and IL-1β levels in a dose-dependent way as compared to Diabetic Parkinson's group receiving no treatment (Fig. 2).

4. Discussion

In the present study, the rat model of Parkinson's disease was induced by MPTP administration which induced Parkinson's disease in all rats treated with it proved by a significant increase in the catalepsy score and a significant decrease in striatal dopamine level. It has been established that MPTP reaches the dopaminergic neurons via the dopamine transporter and triggers their degeneration (Hirsch et al., 2003).

In the Parkinson's group, striatal antioxidant enzyme SOD, CAT and GSH activities have been significantly reduced, with significant increases in striatal MDA, a marker of lipid peroxidation, indicating that oxidative stress involved in the pathogenesis of MPTP-induced Parkinson's disease as reported by Shi et al., (2016), previous study has shown that oxidative stress inhibits the antioxidant protective systems, including SOD, CAT, and GSH, and destroys proteins, lipids, and DNA, eventually resulting in damage to the dopaminergic neurons in the substantia nigra (Maj et al., 2010).

In addition, the levels of proinflammatory cytokines, TNF-α and IL-1β, have increased significantly in the Parkinson group in the present study. Ferger et al., (2004) reported that in the MPTP model of PD, the inflammatory reactions have been determined, and elevated level of the proinflammatory cytokines TNF-α was detected. This can be explained by the excessive microglial activation produced by MPTP, proposing that neuro inflammation and the activated microglia are crucial in PD pathogenesis (Tansey and Goldberg, 2010). Microglia involved in the immune defense and surveillance, and it activates several proinflammatory cytokines, including IL-1β and TNF-α, which can directly induce apoptosis in the dopaminergic neurons (Niranjan 2014).

In order to study the effect of diabetes on the PD-induced biochemical and motor impairments; diabetes induced in the present study by streptozotocin (STZ), a Glucosamine–nitrosourea compound derived from Streptomyces achromogenes. STZ damages the pancreatic β cells, resulting in hypoinsulinemia and hyperglycemia (Lenzen, 2008).

We found that in the Parkinson's group with pre-existing diabetes, there is a significant elevation in the catalepsy score and a significant decrease in striatal dopamine level compared to the Parkinson's group. These results are consistent with Aviles-Olmos et al., (2012) stating that hyperglycemia related to insulin resistance or hypoinsulinaemia, has been shown to decrease the basal striatal dopamine concentrations, this is because insulin receptors are abundantly expressed in the substantia nigra and insulin increases the dopamine transporter mRNA in

Table 2
Catalepsy score in the different studied groups.

Parameter	Control Group	Parkinson's Group	Diabetic Parkinson's Group	Diabetic Parkinson's + Low Dose Exenatide Group	Diabetic Parkinson's + High Dose Exenatide Group
Grid test (time in seconds)	6.9 ± 0.8	29.5 ± 4.5[a]	49.9 ± 5.6[a,b]	30.8 ± 4.8[a,c]	16.2 ± 2.7[a,b,c,d]
Bar test (time in seconds)	7.4 ± 0.9	22.6 ± 3.7[a]	46.3 ± 6.4[a,b]	31.2 ± 3.5[a,c]	17.7 ± 1.8[a,b,c,d]

Values expressed as mean ± SEM. $n = 10$, $P < 0.05$ was considered statistically significant.
[a] Significant from control group.
[b] Significant from Parkinson's group.
[c] Significant from Diabetic Parkinson's group.
[d] Significant from Diabetic Parkinson's + Low Dose Exenatide group.

Table 3
Fasting serum glucose and fasting serum insulin in the different studied groups.

Parameter	Control Group	Parkinson's Group	Diabetic Parkinson's Group	Diabetic Parkinson's + Low Dose Exenatide Group	Diabetic Parkinson's + High Dose Exenatide Group
Fasting serum glucose (mmol/L)	5.1 ± 0.4	5.6 ± 0.5	22.7 ± 2.3[a,b]	5.9 ± 0.9[c]	5.5 ± 0.7[c]
Fasting serum insulin (μIU/ml)	33.7 ± 1.9	34.9 ± 2.1	13.2 ± 1.3[a,b]	35.5 ± 2.3[c]	31.9 ± 3.4[c]

Values expressed as mean ± SEM. $n = 10$, $P < 0.05$ was considered statistically significant.
[a] Significant from control group.
[b] Significant from Parkinson's group.
[c] Significant from Diabetic Parkinson's group.

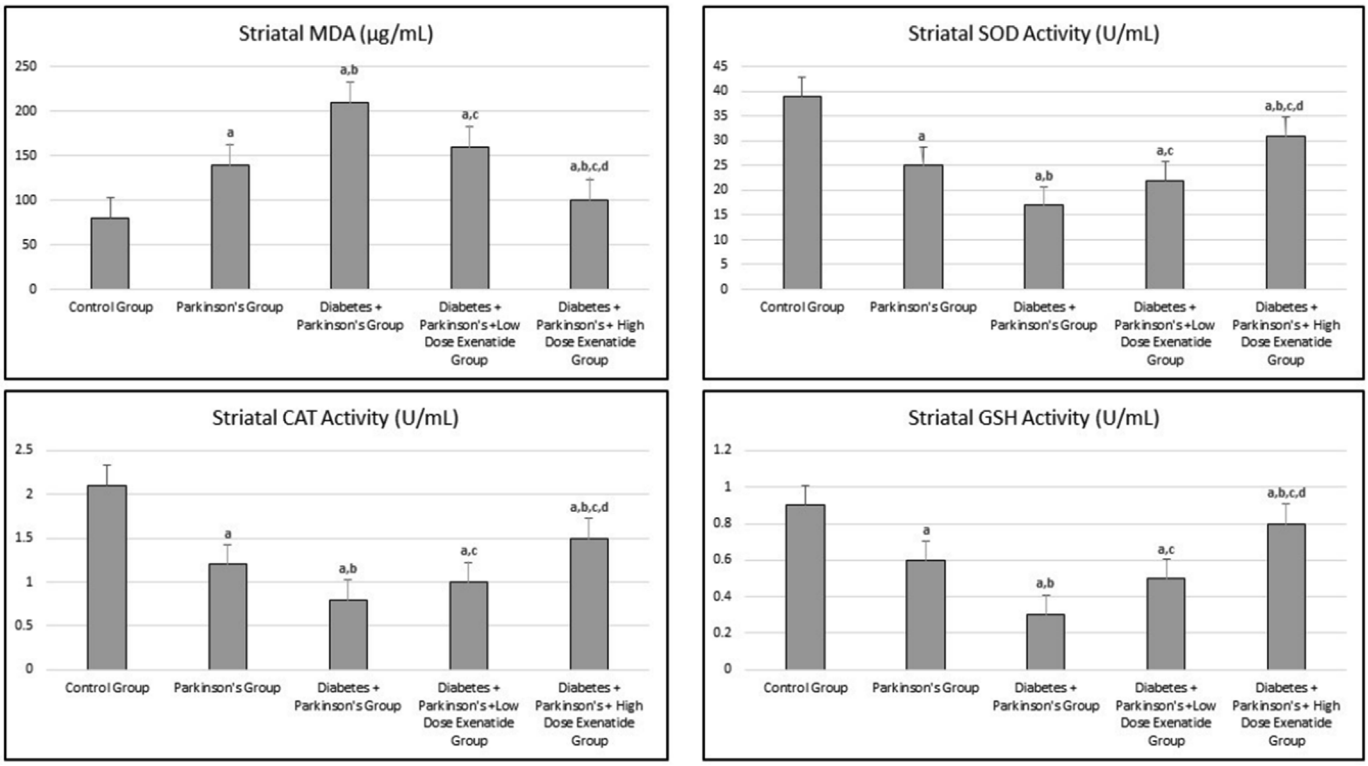

Fig. 1. Striatal oxidative stress markers in the different studied groupsValues expressed as mean ± SEM. $n = 10$, $P < 0.05$ was considered statistically significant
[a] Significant from control group, [b] Significant from Parkinson's group, [c] Significant from Diabetic Parkinson's group, [d] Significant from Diabetic Parkinson's + Low Dose Exenatide group.

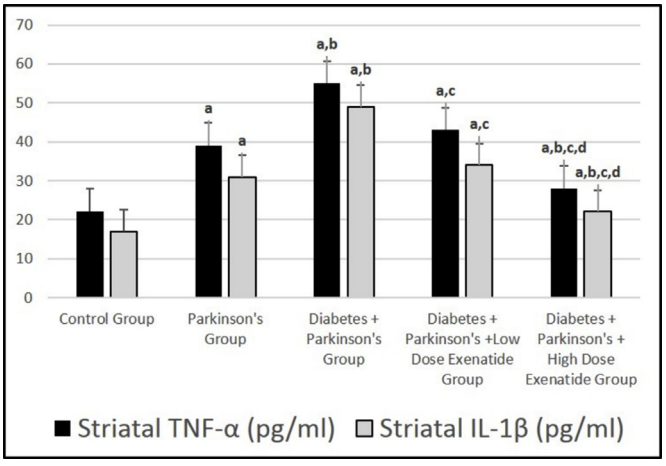

Fig. 2. Striatal inflammatory markers in the different studied groups:
Values expressed as mean ± SEM. $n = 10$, $P < 0.05$ was considered statistically significant
[a] Significant from control group, [b] Significant from Parkinson's group, [c] Significant from Diabetic Parkinson's group, [d] Significant from Diabetic Parkinson's + Low Dose Exenatide group.

substantia nigra, it also regulates brain dopamine concentrations (Cereda et al., 2013).

In the present study, striatal antioxidant enzymes SOD, CAT and GSH activities are significantly reduced in the Parkinson's group with pre-existing diabetes, with significant increases in striatal MDA level compared to the Parkinson's group. These results are consistent with earlier studies that reported that the activities of these antioxidant enzymes decreased in the diabetic brain (Alvarez-Nolting et al., 2012; Miranda et al., 2007). Chronic hyperglycemia triggers oxidative stress with the production of reactive oxygen species, production of reactive oxygen species can also be a mechanism that is underlying dopaminergic cell loss in the hyperglycemic animals (Stranahan and Mattson, 2011).

The brain is mostly susceptible to the oxidative injury because of its high rate of oxygen consumption, excessive production of reactive radicals, and high transition metals levels, such as iron, which catalyze reactive radical production. In addition, neuronal membranes are rich in the polyunsaturated fatty acids which are a lipid peroxidation source (Ansari et al., 2008). Free radicals formed excessively in the diabetes by non-enzymatic proteins glycation, glucose oxidation, and the consequent oxidative degradation of the glycated proteins. Abnormally free radicals high levels, and the concurrent decreasing in antioxidant defense mechanisms may result in damage to the cellular enzymes and organelles, increased lipid peroxidation, and insulin resistance development. These oxidative stress consequences can promote the development of diabetic complications (Rains and Jain, 2011).

Increased in systemic and cerebrovascular inflammation considered as one of the key pathophysiological features in diabetes mellitus and its vascular complications (Goldberg, 2009). Main mechanisms of inflammation caused by hyperglycemia include the neuronal nuclear factor kappa-light-chain-enhancer of the activated B cells (NFκB)-dependent production of proinflammatory cytokines, inflammasome activation, and increased oxidative stress (Lee et al., 2013). These studies can explain the more increase in the levels of the proinflammatory cytokines, TNF-α and IL-1β that found in the present study in the Parkinson's group with preexisting diabetes compared to the Parkinson's group.

Exenatide is a GLP-1 receptors synthetic agonist approved for diabetic treatment with definite useful effects on glucose control, predicted to be mediated by β-cell proliferation, decreased gluconeogenesis, increased insulin production, and weight loss following chronic GLP-1 receptor stimulation in the gastrointestinal tract (Aviles-Olmos et al.,

2013). This explains the exenatide hypoglycemic effect observed in the present study in the exenatide-treated group and its ability to return the diabetic rats' glucose and insulin to normal levels even at their low dose. Exenatide has also been shown to protect the β islet cells from apoptosis, prevent damage to the mitochondrial DNA encoded genes and stimulate the mitochondrial biogenesis (Fan et al., 2010).

GLP-1 receptor expression is commonly found in different cells and organs, including the heart, kidney, lung, endothelial cells, hypothalamus, astrocytes, microglia, and neurons besides pancreatic beta-cells (Arakawa et al., 2010; Fujita et al., 2014; Goke et al., 1995; Iwai et al., 2006; Romani-Perez et al., 2013; Thorens, 1992), indicating that GLP-1 may have additional functions other than the glucose-lowering effects.

The results of the present study demonstrated that treatment of Diabetic Parkinson's group with exenatide significantly improved the striatal dopamine level, and decreased the raised catalepsy score in a dose dependent way as compared to Diabetic Parkinson's group receiving no treatment. These findings are consistent with earlier studies that reported that in a rat model of Parkinson's disease, GLP-1 analogue has demonstrated protection of the substantia nigra dopaminergic neurons and prevention of basal ganglia dopamine loss while retaining motor control (Tan et al., 2009), (Ji et al., 2016), (Chen et al., 2015). Moreover, it was reported that GLP-1 analogue reverses the biochemical and behavioral deficits in a rodent model of Parkinson's disease (Kim et al., 2017; Rampersaud et al., 2012).

As described above, it has been identified that diabetic hyperglycemia has a negative neuronal impact; thus, by its glucose-lowering action, GLP-1 can produce in the neuroprotective effect. Nevertheless, we also investigated whether GLP-1 has a direct neuronal effect by analyzing its impact on striatal inflammatory and oxidative stress status to conclude whether it has its neuroprotective effect only indirectly through its hypoglycemic effect or also through its direct effect on the brain.

GLP-1 agonists can cross the blood–brain barrier (Hunter and Holscher, 2012), so the effect of GLP-1 treatment on cellular pathways involved in inflammation and oxidation investigated. We found that treatment of Diabetic Parkinson's group with exenatide produced a block of the raised level of lipid peroxidation marker, MDA, and the decrease in the activity of antioxidant enzyme, SOD, GSH and CAT, seen in striatal tissue of Diabetic Parkinson's group receiving no treatment in a dose dependent way. Similarly, Muscogiuri et al., (2017), and Spielman et al., (2017) stated that incretins reduce brain oxidative stress by inhibiting the accumulation of intracellular reactive oxygen species (ROS) and the release of nitric oxide (NO) together with increased superoxide dismutase 1 (SOD1) and the antioxidant glutathione peroxidase 1 (GPx 1) expression.

On studying the effect of GLP-1 on striatal inflammatory status, we found that GLP-1 has a definite anti-inflammatory effect, since treatment of Diabetic Parkinson's group with exenatide significantly reduced the striatal TNF-α and IL-1β levels in a dose dependent way as compared to Diabetic Parkinson's group receiving no treatment. GLP-1 receptor stimulation has been reported to attenuate the synthesis of interleukin-1β (IL-1β) pro-inflammatory cytokine in the activated astrocytes (Iwai et al., 2006). In astrocytes, GLP-1 prevented the lipopolysaccharide-induced IL-1β expression by increase of cAMP (Iwai et al., 2006).

From other points of view, the efficacy of GLP-1 in PD models discussed in earlier works; Li et al., (2016) showed that GLP-1 maintained neuronal cell viability and prevented dopaminergic degenerative processes, as well as rotenone-induced apoptotic and neuronal death signaling pathways. This effect can be attributed to the activation of growth factor signaling via the GLP-1 receptor, that inhibits apoptotic signaling (Li et al., 2009). In addition, the GLP-1 receptor induces up regulation of the anti-apoptotic protein B-cell lymphoma 2 (Bcl-2) (Sharma et al., 2014). Bcl-2 acts to reserve mitochondrial integrity by preventing the loss of mitochondrial membrane potential and/or release of pro-apoptotic proteins such as cytochrome C into the cytosol (Harada and Grant, 2003). Thus, this seems that GLP-1 act by improving the course of the pathology rather than just exerting a symptomatic and acute effect. An additional component of the effect mediated by GLP-1 could be a trophic effect on remaining dopaminergic neurons (Bertilsson et al., 2008).

Recently Erbil et al., (2019) study reported that GLP-1 is effective in partially or fully reversing the effects of neuropathological changes related with Alzheimer's disease, Parkinson's disease, neurovascular complications of diabetes, neurotoxic compounds, or vascular occlusion. Possible mechanisms that offer neuroprotection are enhancing the viability of the nerve cells and restoring neurite outgrowth by increased neurotrophic factors, decreasing apoptosis, decreasing the level of pro-inflammatory ingredients, reduced oxidative damage, decreased cerebral edema, and strengthening blood brain barrier.

5. Conclusion

The prominent finding of this study was that GLP-1, in addition to its diabetes treatment effectiveness, exerts significant neuroprotection effects against neuronal damage in PD which worsened with the presence of diabetes. The mechanisms involved could be correlated to its anti-inflammatory and anti-oxidant stress effects through its ability to suppress the inflammatory cytokines expression, and increase the activity of the antioxidant defense enzymes.

The preliminary findings of this study indicate that GLP-1 can exert its neuronal protective effects via a direct effect on the brain independently of controlling the hyperglycemia and the consequent removal of neuronal glucotoxicity effect. This proved by increasing the neuronal GLP-1 effect in a dose dependent manner although the blood glucose return to its normal level with the GLP-1 low dose. These visions afford the opportunity to develop therapeutic approaches directed at specific pathogenic mechanisms, including diabetic control, anti-inflammatory, antioxidant, and insulin-stimulatory drugs that can be effective in protecting against neurodegenerative central nervous system disorders by preventing or stopping the neuronal damage progression in the case of diabetes. However, it is clear that further basic mechanistic and clinical researches required before any potential therapeutic benefits may be realized. Hoping this research will help people with PD, especially those with diabetes, relatively soon.

Limitation

As far as the results of the present study look promising, but longer and larger trials needed to fully test the effects of GLP-1 in diabetic people with Parkinson's since the animal model we used requires the use of chemicals which directly and very quickly harm dopamine-producing nerve cells. This is different to Parkinson's development in people that happen slowly, and affects other types of nerve cells, not only the dopamine-producing ones.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.neuint.2019.104583.

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