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Full Length Research Paper

Nicotinamide: A cytoprotectant against streptozotocin-induced diabetic damage in wistar rat brains

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Nicotinamide is being used in experimental and clinical trials examining the prevention of type-1 diabetes mellitus. However, the precise mechanisms underlying the antidiabetic and neuroprotective effects of nicotinamide require further analysis. Our goals are to evaluate the protective effect and the cellular and molecular mechanisms of nicotinamide against brain damage induced by type-1 diabetes in rats. Type-1 diabetes was induced by i.p injection of streptozotocin (50 mg/kg). Ten days after the induction of diabetes, rats were divided into two groups, control diabetic group and nicotinamide-treated group. Nicotinamide was i.p administered at daily dose of 100 mg/kg for a period of 4 weeks. Another group of normal animals was served as normal control group. The diabetic group showed a significant ($p < 0.05$) decrease in the content of brain DNA, RNA and glutathione, whereas, the contents of lipid peroxide, as malondialdehyde, and nitric oxide were significantly increased. The activities of aldose reductase, sorbitol dehydrogenase and cytochrome oxidase were significantly increased, whereas, the activities of glutathione reductase, glutathione peroxidase, glutathione-S-transferase and superoxide dismutase were significantly decreased. Nicotinamide administration produced restoration of brain malondialdehyde, nitric oxide, glutathione, RNA, DNA levels and the activities of the most measured enzymes. In conclusion, nicotinamide could ameliorate brain damage induced by type-1 diabetes in Wistar rats. The present data provide new approaches for the precise cellular and molecular mechanisms of the neuroprotective effect of nicotinamide.

Key words: Nicotinamide, STZ-diabetes, rat brain, polyol pathway, oxidative status, antioxidant systems, DNA and RNA levels.

INTRODUCTION

The world is facing an explosive increase in the incidence of diabetes mellitus and cost-effective complementary therapies are needed. Diabetes is now considered to be a vascular disease. The cost of treating the micro-vascular component (retinopathy, nephropathy and neuropathy) and controlling the macro-vascular component is a serious drain on health resources particularly in developing countries. The development of diabetes-associated complications in the nervous system was found to be directly attributed to the increased glucose concentration as well as increased polyol pathway activity in brain of diabetic subjects. Many studies showed that hyperglycemia is among the contributing factors involved in most diabetic complications through excessive production of reactive oxygen spices (ROS) (Hunt et al., 1988). Many investigators considered that glucose-induced oxidative-nitrosative stress is critical pathogenic mechanism that initiates a cascade of downstream metabolic and neurovascular perturbations (Hounsom et al*.,* 2001). Insulin administration does not prevent long-term complications of diabetes, as the optimal insulin dosage is difficult to adjust. An alternative strategy is the use of natural agents possessing hypoglycemic effect and can also attenuate the diabetes-induced complications.

A promising agent, nicotinamide, has been shown to protect pancreatic islets from inflammation and stimulate endocrine differentiation of rat β-cells *in vivo* (Banerjee et al., 2005). Nicotinamide is classed as a food additive rather than a drug and has not, therefore, required the

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formal safety evaluation normally expected of a new therapy (Knip et al., 2000). Nicotinamide can act as a weak poly ADP-ribose polymerase (PARP) inhibitor; antioxidant (Melo et al., 2000), improve energy status (Yang et al., 2002) and inhibit apoptosis and cell death in ischemic tissues (Stevens et al., 2007). Additionally, nicotinamide is the precursor for the coenzyme β-nicotinamide adenine dinucleotide (NAD^+) and is considered to be necessary for cellular function and metabolism. Interest in nicotinamide has shifted from its role as a nutrient to that as a novel neuroprotective agent (Maiese and Chong, 2003). Nicotinamide enhances neuronal survival during a variety of insults such as free radical exposure and oxidative stress (Klaidman et al., 2001), since nicotinamide can function as a free radical scavenger (Shen et al., 2004). Nicotinamide, via its PARP inhibitory action, is seemed neuroprotective in pathological conditions associated with oxidative stress (Maiese and Chong, 2003). Moreover, nicotinamide has been used in human clinical trials with low incidence of side effects or toxicity (Gale et al., 2004). However, the effect of nicotinamide on type-1 diabetes-induced brain damage in animal models has not been fully investigated. In the present study, we aimed to evaluate the protective effect and shed more light on the cellular and molecular mechanisms of nicotinamide against brain damage induced by type-1 diabetes in Wistar rats.

MATERIALS AND METHODS

Animals

A total of 36 male albino rats of Wistar strain, obtained from the central animal house of Faculty of Pharmacy, Cairo University, Cairo, Egypt, weighing 170 – 200 g, were used in the present study. All rats were housed in a room with controlled environment, at a constant temperature of 23 ± 1ºC, humidity of 60 ± 10% and a 12 h light/dark cycle. The animals were housed in groups and kept at constant nutritional conditions throughout the experimental period. The experimental protocols were approved by the Ethical Committee of Cairo University.

Chemicals and drugs: Streptozotocin (STZ), nicotinamide; enzymes, coenzymes and fine chemicals were obtained from Sigma-aldrich Co. (St. Louis, MD, USA). Other chemicals are of Analytical grade.

Experimental groups and protocol: Type-1 diabetes was induced in fed rats by i.p injection of STZ (50 mg/kg) freshly prepared in 0.1 M sodium citrate buffer, pH 4.5 (Wahieb and Godin, 1987). During the first 24 h of diabetes induction, STZ-treated animals were allowed to drink 5% glucose solution to overcome drug-induced hypoglycemia (Kakkar et al., 1997). Forty-eight hours after STZ administration, diabetes was confirmed by the presence of hyperglycemia and glucosuria. STZ-treated animals showed blood glucose less than 400mg/dl and glucosuria lower than (+3) were discarded. Body weight of each animal was monitored weekly during the period of the study. Only STZ-treated rats that showed weight loss, or no-weight gain, were used in the study. This effectively limited the experimental variation due to different degrees of the disease (Parinandi et al., 1990). Ten days after the induction of diabetes, STZ-treated rats were divided into two groups, each containing 12 - 14 animals. The first group (14 rats) was left without treatment till the end of the experimental period and served as control diabetic group. The second group (12 rats) was i.p treated with nicotinamide, at daily dose of 100 mg/kg for a period of 4 weeks, and served as nicotinamide-treated group. Another group of normal animals (10 rats) was received citrate buffer by the same dose similarly as the control diabetic group and served as normal control group. Treated and control animals were allowed free access to water and kept at constant environmental and nutritional conditions throughout the experimental period.

Blood sampling: At the end of the experimental period, all animals were sacrificed. Portion of blood was collected in heparinized tubes and centrifuged at 600 xg for 15 min. The separated plasma was used for glucose determination according to the method of Burrin and Price (1985), by using the reagents of kit supplied by Sera-Pak of Bayer (France). Another portion of blood was collected; centrifuged at 600 x g for 15 min and the separated serum was used for the determination of fructosamine concentration according to the method of Schleicher and Vogt (1990), by using the kit supplied by Quimica Clinica Aplicada (Spain). The remainder of the blood sample was collected in citrated tubes and used for glycated Hb estimation (Abraham et al., 1978), using the kit provided by Stanbio Laboratory, USA.

Tissue sampling: The skulls were split on ice and salt mixture and the whole brains were removed; rinsed with ice-cold saline; blotted dry and frozen rapidly using a mixture of $CaCl_2$; NaCl and ice (-55°C). The frozen tissue was powdered; mixed and accurately weighed amounts were treated differently for the separation and estimation of the studied parameters. Two portions of brain powder were used to prepare 10% homogenate in 1.15% KCl and 5% homogenate in 3% sulfosalicylic acid and centrifuged at 1000 xg at 4°C for 20 min. The resulted supernatants were used for the assay of brain malondialdehyde (MDA) (Yoshioka et al., 1979) and glutathione (GSH) (Srivastava and Beutler, 1968), respectively. Portion of brain powder was homogenized in Tris-sucrose buffer, pH 7.4 (5% homogenate) and centrifuged at 2000 xg at 4°C for 10 min. The resulted supernatant was used for estimating nitric oxide (NO) concentration, as nitrate and nitrite by the Griess reaction (Harold et al., 1992), and cytochrome oxidase activity, by the method described by Smith (1955), in which the decrease in absorbency due to the oxidation of reduced cytochrome c by cytochrome oxidase, was measured spectrophotometrically at 550 nm. A unit of cytochrome oxidase activity is defined as the amount of enzyme that oxidizes 1 µmole ferrocytochrome c per minute. Another portion of brain powder was homogenized in Tris-sucrose buffer, pH 7.4 (10% homogenate) and centrifuge at 105,000 xg at 4°C for 30 min. The separated cytosolic fraction was used for estimating the activities of the following enzymes: (i) Glutathione-S-transferase (GST), depending on the ability of GST to catalyze the formation of glutathione adduct with 1-chloro,2,4 dinitrobenzene which can be measured by noting the net increase in absorbance at 340 nm (Habig et al., 1974). One unit of GST was defined as the amount of the enzyme catalyzing the formation of 1µmol of the conjugated product (Vodela and Dalvi, 1997). (ii) Glutathione reductase (GR), depending on the reduction of oxidized glutathione using NADPH, in the presence of glutathione reductase, which can be followed spectrophotometrically by measuring the decrease in the absorbance at 340 nm for 3 min due to the oxidation of NADPH to NADP (Long and Carson, 1961).(iii) Glutathione peroxidase (GPX), depending on that, the of oxidation of glutathione by hydrogen peroxide, in the presence of glutathione peroxidase, was determined by measuring the rate of oxidized glutathione formation,

Table 1. Effects of nicotinamide treatment on plasma glucose; serum fructosamine contents and blood located Hb % in diabetic rats.

Group Parameters	Normal	Diabetic	Nicotinamide treated
Plasma glucose mg/dL	94.62 ± 9.79	a 901.78 ± 26.41	a,b 371.70 ± 33.94
Serum Fructosamine □ Mole / L	224.65 ± 10.89	a 360.84 ± 23.47	300.37 ± 21.29
Blood Glycated Hb %	6.72 ± 0.91	a 21.65 ± 1.09	a,b 11.32 ± 1.25

Results expressed as Means ± SEM.
(a) Significantly different from normal control at $p < 0.05$.
(b) Significantly different from diabetic control at $p < 0.05$

Table 2. Effect of nicotinamide treatment on AR and SD activities in diabetic rat brains

Group Parameters	Normal	Diabetic	Nicotinamide treated
ARnmoles NADPH/mg protein/hr	27.5 ± 2.13	**a** 66.2 ± 3.55	**b** 22.5 ± 1.61
SD nmoles NADH/mg protein/min	35.6 ± 1.99	**a** 52.4 ± 2.47	**b** 36.1 ± 3.06

Results expressed as Means ± SEM.
a). Significantly different from normal control at $p < 0.05$.
b). Significantly different from diabetic control at $p < 0.05$.

Table 3. Effect of nicotinamide treatment on MDA and NO contents in diabetic rat brains.

GroupParameters	Normal	Diabetic	Nicotinamide treated
MDA nmoles/gm t wt	40.2 ± 3.40	**a** 83.9 ± 4.98	**b** 54.9 ± 3.213
NO nmoles/gm t wt	16.3 ± 1.48	**a** 43.1 ± 2.64	**b** 20.5 ± 1.06

Results expressed as Means ± SEM.
(a) Significantly different from normal control at $p < 0.05$.
(b) Significantly different from diabetic control at $p < 0.05$.

this in turn was determined by following up the decrease in absorbance of NADPH at 340 nm (Arthur and Boyne, 1985). Also, superoxide dismutase (SOD) was estimated according to the method of Marklund and Marlund (1974). In this method, the inhibition of pyrogallol autoxidation, brought about by SOD, can be employed in a rapid and convenient method for the determination of the enzyme. Another portion of brain powder was homogenized in potassium phosphate buffer, pH 7 (20% homogenate). Part of the resulted homogenate was centrifuged at 105,000 xg for 45 min at 4°C and used for estimating aldose reductase (AR) activity, depending on the reaction between D-glucose and NADPH.H+, in the presence of AR. The decrease in the absorbance due to the oxidation of $NADPH.H^+$ to $NADP^+$ was followed spectrophotometrically at 340 nm (Chauncey et al., 1988). The remainder of the homogenate was centrifuged at 30,000 x g for 30 min at 4°C and the resulted supernatant was used for estimating sorbitol dehydrogenase (SD) activity, depending on the reaction between D-sorbitol and NAD+, in the presence of SD. The increase in the absorbance due to the reduction of NAD+ to NADH was followed spectrophotometrically at 340 nm (Leissing and McGuinness, 1982). Another portion of brain powder was homogenized in 0.25 M sucrose-TKM buffer (0.05M Tris-HCl, 0.025M KCl and 0.005M $MgCl_2$), pH 7.5, to prepare 15% homogenate. Perchloric acid (0.1 ml of 0.3 M) was added; stand at 0°C for 15 min; centrifuged at 2000 x g at 4°C for 10 min and the resulted precipitate was used for estimating DNA (Giles and Myers, 1965) and RNA (Blobel and Potter, 1968) contents. The protein concentration of the above supernatants was estimated by the method described by Lowry et al (1951).

Statistical analysis: The results are given as the Mean ± SEM. Comparison between the different groups were carried out by One Way Analysis Of Variance (ANOVA), followed by Kruskal-Wallis test. $P < 0.05$ was considered significant.

RESULTS

STZ-diabetes caused significant elevation in plasma glucose; serum fructosamine concentrations and blood glycated Hb%, reaching to about 983, 173 and 341% of the normal values, respectively. Nicotinamide treatment produced significant decrease in plasma glucose; serum fructosamine concentrations and blood glycated Hb%, reaching to 47, 50 and 52.3% of the diabetic values, respectively (Table 1). STZ-diabetes produced significant elevation in brain AR and SD activities, reaching to about 241 and 147% of the normal values, respectively. Nicotinamide normalized the activity of both enzymes (Table 2). As shown in Table 3, STZ-diabetes caused significant elevation in brain MDA and NO concentrations, reaching to 209 and 264% of the normal values, respectively. Ni-

Table 4. Effect of nicotinamide treatment on GSH content, GR; GPX; GST and SOD activities in diabetic rat brains.

Group Parameters	Normal	Diabetic	Nicotinamide treated
GSH μg/gm t wt	463 ± 11.7	**a** 214 ± 16.1	**b** 488 ± 18.1
GR U/mg protein	121 ± 4.57	**a** 87.6 ± 3.48	**b** 106±6.03
GPXU/mgprotein	79.8 ± 4.32	**a** 59.4 ± 4.71	**b** 83.1 ± 5.93
GSTU/mgprotein	69.3 ± 1.5	**a** 53.8 ± 3.14	**b** 64.0 ± 4.04
SODU/mgprotein	145.4 ± 4.63	**a** 88.2 ± 5.98	**b** 131 ± 10.9

Results expressed as Means ± SEM.
(a) Significantly different from normal control at $p < 0.05$.
(b) Significantly different from diabetic control at $p < 0.05$.

Table 5. Effect of nicotinamide treatment on cytochrome oxidase activity, DNA and RNA contents in diabetic rat brains.

Group Parameters	Normal	Diabetic	Nicotinamide treated
Cytochrome oxidase n moles reduced cytochrome c/ mg protein/ hr	10.4 ± 0.367	**a** 109.7 ± 8.338	**a** 113 ± 1.29
DNA mg/gm t. wt.	10.39 ± 0.69	**a** 4.45 ± 0.185	**b** 9.28 ± 0.837
RNA mg/gm t. wt.	1.092 ± 0.067	**a** 0.72 ± 0.035	**b** 1.16 ± 0.097

Results expressed as Means ± SEM.
(a) Significantly different from normal control at $p < 0.05$.
(b) Significantly different from diabetic control at $p < 0.05$.

Nicotinamide normalized these parameters. Table 4 illustrated that STZ-diabetes, significantly, suppresses the studied antioxidant systems, GSH concentration; GR; GPX; GST and SOD activities, reaching to about 46, 73, 74, 78 and 61% of normal values, respectively. Nicotinamide restored these antioxidant parameters. STZ-diabetes produced marked elevation in brain cytochrome oxidase activity reaching to about 1054%, along with significant decrease in brain DNA and RNA levels, reaching to about 43 and 66% of the normal values, respectively. Nicotinamide normalized DNA and RNA levels (Table 5).

DISCCUSION

The current data showed that STZ-induced diabetes was accompanied by development of hyperglycemia; increased serum fructosamine concentration and blood glycated Hb%. Increased hepatic glucose production plus decreased hepatic glycogen synthesis and glycolysis are the major mechanisms in diabetes mellitus that result in hyperglycemia (Guignot and Mithieux, 1999). Previous studies revealed that high blood glucose-induced deterioration of pancreatic β cells may be due to oxidative stress, thus, antioxidants have beneficial effects on pancreatic β cells by neutralizing such conditions (Kaneto et al., 2001). Nicotinamide administration significantly lowered the biomarkers of hyperglycemia especially blood glycated Hb % which is considered the index of long term diabetic control. The hypoglycemic effect of nicotinamide seems to be mediated by modulating the hepatic glucose regulating enzymes. Previous study revealed that nicotinamide, which is involved in NAD^+ biosynthesis, cause decrease in blood glucose content; stabilize the content of 2,3 DPG and of glycated Hb in RBCs. Nicotinamide can decrease the rate of Hb glycosylation and enhance tissue O_2 utilization under hypoxic conditions (Velikii et al., 1995).

Rats subjected to STZ-diabetes showed significant activation of polyol pathway regulating enzymes, aldose reductase (AR) and sorbitol dehydrogenase (SD). The reported development of diabetes-induced complications in brain of diabetic subjects is directly attributed to the increased polyol pathway activity. The accumulated sorbitol, in nervous tissue of diabetic animals, can increase the cellular osmolarity, resulting in water retention; cell oedema and increases in cytosolic Na^+ concentration. All may contribute to the etiology of diabetic neuropathy (Nattrass, 1986). The current increase in the activity of brain AR could be attributed mainly to the marked elevation in brain glucose content demonstrated previously in diabetic rat**s** (Kador et al., 1985). Schwann cells of diabetic rats show abundantly expressed AR which results in the development of both

axonopathy and sensory neuropathy (Calcutt et al., 2004). Also, AR-containing schwann cells were reported to be the main source of ROS and reactive nitrogen species in the nervous system of the diabetic subjects (Obrosova et al., 2005). The increased brain SD activity, shown in STZ-diabetic animals, might be considered as an enzymatic adaptation which facilitates the degradation of the reported accumulated sorbitol. Over expression of SD stimulates ROS generation in high glucose-exposed retinal pericytes, and subsequently, potentiates the cytopathic effects of glucose (Amano et al., 2002). Interestingly, nicotinamide administration could attenuate, significantly, the active polyol pathway regulating enzymes shown in the brains of the diabetic group. Nicotinamide, as a precursor of NAD+ biosynthesis, can increase the free NADP+/NADPH with subsequent inhibittion of AR, a NADPH-dependent enzyme. In addition, nicotinamide can decrease the high brain glucose content, the main initiator of AR enzyme, via increasing the utilization and decreasing the synthesis of glucose through gluconeogenesis. Additionally, suppression of PARP activity by nicotinamide decreases the consumption of NAD^+, the substrate of PARP, leading to increased blood and liver NAD^+ concentrations (ApSimon et al., 1995). Increase in the free NAD^+/NADH ratio was reported to be accompanied by inhibition of the key gluconeogenic enzymes (Velikii et al., 1992). Also, nicotinamide could normalize brain SD activity which could be considered as a net result of two opposite effects, the stimulatory action of the increased NAD^+ level, produced by nicotinamide, and the inhibitory action exerted by the elevated brain GSH level, shown in the nicotinamide-treated group. Increased cellular GSH level was reported to be one of the inhibitory factors for SD activity (Bergmeyer et al., 1988). Thus, the present study provides an evidence for a new molecular mechanism concerning with the antidiabetogenic and the neuroprotective efficiencies of nicotinamide. This is achieved via its inhibitory action on the brain polyol pathway, a main contributing factor for diabetes-induced brain damage.

In the present study, rats subjected to STZ-diabetes showed significant increase in brain biomarkers of oxidative stress, MDA and NO contents. Oxidative stress has emerged as a critical factor in the development of chronic diabetic complications **(**Cameron et al., 1994). Increased oxygen free radicals activity in diabetes has been ascribed to glucose-protein interactions and to glucose-induced activation of AR pathway (Fondelli et al., 1993). Many investigators considered that glucose-induced oxidative-nitrosative stress is critical pathogenic mechanism that initiates a cascade of downstream metabolic and neurovascular perturbations (Hounsom et al., 2001). Additionally, activation of the nuclear PARP, demonstrated previously in diabetes mellitus**,** is now viewed as an important effector of oxidative – nitrosative injury (Obrosova et al., 2004), which results in cleavage of NAD^+ and the formation of nicotinamide and ADP-ribose residues attached to nuclear and extranuclear proteins (Southan and Szabo, 2003). PARP activation leads to the depletion of its substrate NAD^+; energy failure; inhibition of glyceraldehyde 3-phosphate dehydrogenase of glycollysis; altered gene transcription, and, in extreme cases, cell necrosis (Garcia et al., 2001). Administration of nicotinamide to diabetic rats could normalize brain level of both oxidative biomarkers. The properties of nicotinamide, which functions as both a free radical scavenger (Shen et al., 2004) and PARP inhibitor, would make it an attractive agent to combat these pathogenic pathways. Nicotinamide, via its PARP inhibitory action, is seemed to be as neuroprotective in pathological conditions associated with oxidative stress (Maiese and Chong, 2003). Moreover, it was reported that nicotinamide used in human clinical trials showed low incidence of side effects or toxicity (Gale et al., 2004).

The present results demonstrated that STZ-diabetes significantly suppresses the measured antioxidant systems including GSH content; GR, GPX, GST and SOD activities. Treatment of the diabetic rats with nicotinamide could improve all the studied antioxidant systems**.** The high flux of glucose through the activated polyol pathway in diabetic brain may consume NADPH, which results in decreased level of reduced GSH, and consequent decrease in the activities of all GSH-dependent enzymes like GR; GPX and GST. Previously, it was demonstrated that, in rat primary cortical neurons, nicotinamide reduce LDH release; ROS production; calcium influx; caspase-3 activation and cell injury after oxygen-glucose deprivation and re-oxygenation (Shen et al., 2004). Also, it was reported that, in neurons exposed to high glucose, nicotinamide reduces superoxide and hydrogen peroxide production and counteracts glucose-mediated (or hydrogen peroxide-induced) cell death (Vincent et al., 2005). Earlier evidences suggested that nicotinamide can exert a direct neuroprotective effect in both neurons and glial cells of diabetic subjects (Veres et al., 2004).

Our data revealed that, in diabetic rat brains, there is marked decrease in DNA and RNA contents, together with marked elevation in cytochrome oxidase activity. Diabetes-induced oxidative stress can contribute to cellular damage and appears to be the common apoptotic mediator, most likely via lipid peroxidation (Buttke and Sandstrom, 1994). Enhancement of ROS generation has been reported to elicit translocation of cytosolic bax to mitochondria and to activate bax to induce the release of cytochrome c from mitochondria. This investigation might be in relation with the present data that diabetic rat brains showed marked elevation in cytochrome oxidase activity. Diabetes-induced PARP activation has been shown to be a consequence of the increased production o f ROS and

reactive nitrogen species, secondary to DNA single-strand breakage, thereby resulting in a self-perpetuating cycle (Obrosova et al., 2004). The extracellular signal-regulated kinase and p38 mitogen-activated protein kinase functioned as transducers for the detrimental effects of hyperglycemia by promoting neurodegeneration (Chong et al., 2002). These signal transducers can be activated by glucose and oxidative stress via a mechanism involving PARP (Veres et al., 2004). In the current study, treatment of diabetic rats with nicotinamide could improve such depletion in brain DNA and RNA contents. Interest in nicotinamide has shifted from its role as a nutrient to that as a novel neuroprotective agent (Maiese and Chong, 2003). Nicotinamide can enhance neuronal survival during variety of insults such as free radical exposure and oxidative stress (Garcia et al., 2001; Southan and Szabo, 2003). The mechanism of nicotinamide-mediated neuroprotection may be in part due to inhibition of both caspase-3 and the release of cytochrome c from mitochondria during oxygen–glucose deprivation (Chong et al., 2004). Neuroprotection by nicotinamide was investigated to function at the mitochondrial level**.** It was shown that, nicotinamide, *in vivo*, prevents ethanol-induced cytochrome c release and the following caspase-3 activation (Maiese and Chong, 2003). Although the determination of the precise mechanisms underlying the neuroprotective effects of nicotinamide requires further analysis, it is possible that nicotinamide may function in stabilizing the cellular energy metabolism. During an oxidative stress, depletion of NAD^+ is considered a critical factor in precipitating cell death due to compromised energy supply (Beal, 2000). Nicotinamide can increase the amount of NAD^+ in the brain, and, thus, preventing its depletion and consequent energetic decline (Yang et al., 2002). This is in consistent with a study that increasing NAD^+ biosynthesis prevented axonal degeneration (Araki et al., 2004). In addition, the properties of nicotinamide that can function as both a free radical scavenger and PARP inhibitor would make it an attractive agent to combat these pathogenic pathways. However, nicotinamide does not directly alter the activity of either p38 or c-Jun N-terminal kinase (Chong et al., 2004), which suggests that its effects may be more distal, such as preventing the activation of caspase-1 and caspase-3.

Conclusion

Nicotinamide can successfully ameliorate brain damage induced by type-1 diabetes in Wistar rats as evidenced in the present study by: (i) Nicotinamide can significantly lower the biomarkers of hyperglycemia**.** (ii) Nicotinamide exerts inhibitory action on the brain polyol pathway. (iii) Nicotinamide can normalize brain levels of both oxidative and nitrosative stress biomarkers, MDA and NO contents (iv) Nicotinamide can improve the studied antioxidant systems**,** GSH level; GR; GPX; GST and SOD activities. (V) Nicotinamide can improve brain DNA as well as RNA levels.

Recommendation

The determination of the precise mechanisms underlying the antidiabetic and neuroprotective effects of nicotinamide requires further analysis. However, in the light of the present study, we may recommend to use nicotinamide as antidiabetic and neuroprotective supplement, since, nicotinamide can function as: anti-hyperglycemic; polyol pathway inhibitory; anti-oxidative-nitosative and neuronal survival-enhancing agent.

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