

CHINESE MEDICAL SCIENCES JOURNAL

REVIEW

C-peptide and Diabetic Encephalopathy

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Key words: C-peptide; diabetes; encephalopathy

With the changes of life style, diabetes and its complications have become a major cause of morbidity and mortality. It is reasonable to anticipate a continued rise in the incidence of diabetes and its complications along with the aging of the population, increase in adult obesity rate, and other risk factors. Diabetic encephalopathy is one of the severe microvascular complications of diabetes, characterized by impaired cognitive functions, and electrophysiological, neurochemical, and structural abnormalities. It may involve direct neuronal damage caused by intracellular glucose. However, the pathogenesis of this disease is complex and its diagnosis is not very clear. Previous researches have suggested that chronic metabolic alterations, vascular changes, and neuronal apoptosis may play important roles in neuronal loss and damaged cognitive functions. Multiple factors are responsible for neuronal apoptosis, such as disturbed insulin growth factor (IGF) system, hyperglycemia, and the aging process. Recent data suggest that insulin/C-peptide deficiency may exert a primary and key effect in diabetic encephalopathy. Administration of C-peptide partially improves the condition of the IGF system in the brain and prevents neuronal apoptosis in the hippocampus of diabetic patients. Those findings provide a basis for application of C-peptide as a potentially effective therapy for diabetes and diabetic encephalopathy.

Chin Med Sci J 2011; 26(2):119-125

AS a common metabolic disorder, diabetes could extensively damage many tissues and organs including heart, retina, kidney, blood vessels, and the nervous system. The incidence of diabetes and its complications is rapidly increasing worldwide. Approximately half of the patients with diabetes die prematurely of a cardiovascular cause and approximately 10% die of renal failure and cerebral complication.¹ Although the pathogenesis of diabetes is not clear yet, the

combined effects of ageing, obesity, and physical inactivity may play key roles in the development of this disease.

Recent studies suggest the association of diabetes mellitus with gradually developing end-organ injury to the central nervous system (CNS). This complication of both type 1 and type 2 diabetes can be referred to as "diabetic encephalopathy". The mechanisms are only partially understood. Both microvascular and macrovascular cerebral diseases occurring in diabetes and the direct neuronal damage caused by chronically elevated intracellular glucose concentration are implicated in encephalopathy. However, it remains unclear to what extent the neuronal impairment is induced directly by elevated intracellular

glucose level. Emerging evidence indicates that the changes of C-peptide level, increased oxidative stress and consequent oxidative damage observed in hyperglycemic conditions and diabetic encephalopathy begin in the mitochondria, which is the major site of reactive oxygen species production.^{2,3} Many studies offer fresh evidence supporting that apart from the vascular dependent mechanisms of brain dysfunction and oxidative and nitrosative stress, C-peptide also exerts a pivotal role in diabetic complications including encephalopathy.⁴⁻⁷

In this review, we will describe the encephalopathy occurring in diabetes and discuss its several possible mechanisms, focusing on C-peptide deficiency and CNS dysfunction.

CLINICAL MANIFESTATIONS OF DIABETIC ENCEPHALOPATHY

Cognitive and neurobehavioral deficits in diabetic encephalopathy

Diabetic encephalopathy is characterized by damaged cognitive functions and neurobehavioral deficits.⁸ Impairments of this kind in both type 1 and type 2 diabetic patients will affect the process of learning, memory, problem solving, informative proposal, mental and motor speed. In addition, specific cognitive disturbances associated with diabetes have recently been described.⁹ Deficits have been repeatedly observed in particular cognitive domains, including memory and psychomotor speed. There is some overlap in the findings of cognitive changes in type 1 diabetes and type 2 diabetes, but an increased risk for dementia or mild cognitive impairment in the latter type. Diabetic patients suffer from significantly more introverted symptoms than their healthy counterparts, especially somatic symptoms, sleeping disturbance, compulsions, and depressive moods.¹⁰

Cerebral stroke in diabetic encephalopathy

In diabetic encephalopathy, cerebral stroke is one of the major causes of death and disability. Eighty percent of cerebral strokes are ischemic, including thrombotic and embolic stroke. The damage could lead to stroke syndromes including vertigo, nystagmus, sensory loss, anopia, facial numbness, amnesia, color anomia, urinary incontinence, or coma, depending on the arterial territory involved. Cerebral stroke of diabetic encephalopathy varies in infarction mechanism and size, ranging from the lacunar strokes of small vessel disease to the large wedge-shaped cortical and subcortical infarctions of embolic stroke. There has been evidence indicating that the severity of cerebral

ischemia depends on the duration as well as the severity of diabetes mellitus.¹¹ In patients with different types of acute ischemic stroke, the prevalence of diabetes is very high in those with small-vessel cerebrovascular accidents and large-artery atherosclerosis, while less common in those with other combined etiologies of stroke. Previous studies show that chronic hyperglycemia, long duration, and insulin resistance may be responsible for an elevated risk of cerebral stroke in diabetic encephalopathy.¹²

Brain changes in diabetes

Neuroimaging can correlate structural brain parameters with cognitive changes. Brain scans can be applied in quantitative monitoring of a disease over time in an individual with 3-dimensional images. Furthermore, magnetic resonance imaging (MRI) can assess suspected involvement of neuroanatomical areas in the diabetic brain, which is problematic to study routinely in tissue-based neuropathology. For these reasons, neuroimaging studies are practically tailor-made for assessment of the cerebral neuropathology of diabetes. van Harten et al¹³ conducted a meta-analysis covering over 40 studies using MRI and/or CT on at least 20 diabetic patients. They identified three common structural changes in the diabetic patients' brains. One is white matter lesions, as some studies showed a positive correlation between the presence of white matter lesions and type 2 diabetes. Another common change is lacunar infarcts. A significant association was found between the presence of lacunar infarct and type 2 diabetes across different study designs. The third one is cortical atrophy. Despite the different methodology, the majority of the publications in the meta-analysis showed a positive correlation between cortical atrophy and type 1 diabetes.

Of the three changes described above, the two least understood are white matter lesions and lacunar infarcts. Technology that probes both functional and structural parameters could offer more insights into the brain changes in diabetes.

Manifestations of diabetic encephalopathy and its causes

Although it is well-known that hypoglycemia can cause brain damages, it remains controversial that whether the above CNS abnormalities of diabetic encephalopathy are induced by diabetes per se or by hypoglycemic episodes, which occur frequently in type 1 diabetic patients receiving intensive insulin treatment. It has been reported that a duration-dependent decline in cognitive function occurs in type 1 diabetic patients without hypoglycemic episodes,^{14,15} and impaired intellectual and cognitive development in

type 1 diabetic children who have not experienced hypoglycemic episodes.¹⁶ The latter impairments correlate with diagnosis at young age and metabolic status at diagnosis. Therefore, encephalopathy may be impacted by diabetes itself, not hypoglycemic episodes in diabetes. Although cerebrovascular disease is barely mentioned in type 1 diabetes, a recent analysis of mortality from cerebrovascular disease in over 20 thousand patients with type 1 diabetes has indicated for the first time that cerebrovascular mortality is raised at all ages in this patient group.¹⁷ Thus, type 1 diabetes is a high risk factor for cerebrovascular mortality.

Neuropsychological researches in type 2 diabetic patients have produced more consistent results than in type 1 diabetic patients. Cognitive deficits and poor performance in abstract reasoning and complex psychomotor functioning occur in type 2 diabetes. Complex cognitive tasks requiring storage and retrieval of new information are influenced, whereas less demanding performances such as immediate memory recall and simple reaction time are not obviously affected.¹⁸ However, no significant impairments in learning, memory, and problem solving skills were found in middle-aged type 2 diabetic patients compared with controls.¹⁹ A review of related studies showed that learning and memory dysfunction is more prominent in elderly type 2 diabetic subjects.²⁰ It may be due to a potentiation of the normal aging process by superimposed diabetes, to the duration of diabetes, or to the combined effect of both factors. The etiology of cognitive dysfunction in type 2 diabetes is not fully understood. A possible explanation is the interaction between metabolic abnormalities intrinsic to diabetes and diabetes-specific complications. After all, the higher frequency of cerebral stroke in type 2 diabetes may be associated with cognitive deficits.^{21,22} A study

analyzing over 5000 type 2 diabetic patients with a novel mathematical model confirmed the higher risk of stroke in type 2 diabetes compared with healthy population.²³ Besides, the neurobehavioral and neuropsychological changes of diabetic encephalopathy are similar to those of the aging cerebra, indicating that diabetic encephalopathy may accelerate the aging of cerebra.²³

Based on these reports mentioned above, the future studies on cerebral stroke in diabetes will provide useful information about the possible underlying mechanisms of diabetic encephalopathy.

POSSIBLE MECHANISMS OF DIABETIC ENCEPHALOPATHY

It is generally considered that four major pathomechanisms lead to the occurrence and development of complications including encephalopathy in diabetes: the polyol pathway and enhanced polyol pathway flux; increased hexosamine pathway throughput and subsequent overmodification of proteins by N-acetylglucosamine; hyperglycemia-provoked activation of protein kinase C isoforms; and increased formation of advanced glycation end-products.²⁴ These pathomechanisms mentioned above have long been regarded as independent processes, or at least their interrelationship was indistinct. However, subsequent studies have proved that these processes are actually linked together and have a common hyperglycemia (and also fatty acid)-dependent cause.²⁴ In spite of these findings, the precise and specific mechanisms underlying neuronal apoptosis and diabetic encephalopathy are not explicit. We presumed that the factors responsible are insulin deficiency with concomitant C-peptide changes, hyperglycemia, and possibly the aging process (Fig. 1).

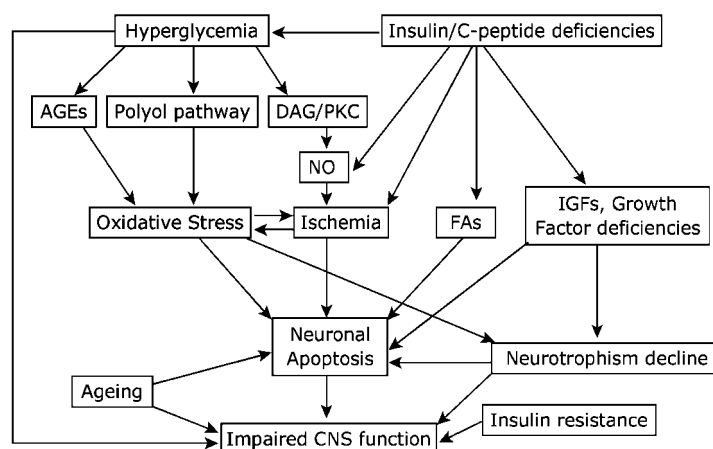


Figure.1 Possible pathways resulting in diabetic encephalopathy involving the central nervous system (CNS) dysfunction.

AGEs, advanced glycation end-products; DAG, diacylglycerol; PKC, protein kinase C; NO, nitric oxide; FAs, fatty acids; IGFs, insulin-like growth factors.

INSULIN/C-PEPTIDE DEFICIENCY

Impact of insulin level on CNS

Since decades ago, many studies have discussed complications of diabetes on the peripheral nervous system, but the influence on CNS has been examined only in the last 10-15 years, although the latter consequence is no less important.²⁵ There has been evidence suggesting a direct link between insulin level and cognitive performance.²⁶ The risk of neurodegeneration and cognitive decline is increased in insulin-resistant patients who do not display hyperglycemia (pre-diabetes). High insulin level may inhibit neuron firing and decrease the activity of choline acetyltransferase, which is involved in forming acetylcholine, a neurotransmitter regulating memory and learning. Insulin is also implicated in the action of other neurotransmitters, such as the GABA_A receptor-mediated synaptic transmission.²⁷

C-peptide is cleaved evenly into halves in the conversion of proinsulin to insulin. Its half-life in the circulation is 2 to 5 times longer than that of insulin, therefore could serve as a stable indicator of insulin secretion. Okereke et al²⁸ measured the C-peptide level in 718 non-diabetic women in their early 60s, and estimated their cognitive functions 10 years later based on telephone interviews. They found significantly worse cognitive functions in those with higher C-peptide level than the cognitive functions in those with lower C-peptide level. Their finding implies that insulin plays an important role in the regulation of brain metabolism. It has also been demonstrated that insulin exerts additional modulatory effects on brain functions such as learning and memory.²⁹ Administration of insulin in cerebral stroke prevents neuronal death and reduces neurological disability. The neuroprotective effects of insulin may be associated with restoration of protein synthesis *via* increased dephosphorylation of eukaryotic initiation factor 2 α . An *in vitro* study demonstrate that insulin has an antiapoptotic effect.³⁰ Sima et al³¹ reported that the insulin receptor is down-regulated in the brain of type 1 diabetic BB/Wor rats, suggesting that impaired insulin action induced by declined insulin receptor expression as well as insulinopenia may play a significant role in CNS apoptosis in diabetes.

C-peptide deficiency in diabetes

In addition to insulin deficiency, C-peptide deficiency has been considered as a pathogenic factor in complications of type 1 diabetes.¹⁴ C-peptide is a cleavage product of insulin synthesis and secreted together with insulin by pancreatic beta-cells upon glucose stimulation. Thus, patients with

impaired insulin synthesis will also become C-peptide-deficient.³² Preclinical and clinical studies suggest that lack of C-peptide may exacerbate diabetes-associated complications including diabetic encephalopathy. Hanafusa et al³³ reported that apart from instability of glucose levels, lack of C-peptide itself might play a role in the development of microangiopathy in fulminant type 1 diabetic patients. Kim et al³⁴ found that patients with lower delta C-peptide (postprandial-fasting C-peptide) quartile had significantly higher prevalences of diabetic retinopathy, nephropathy, and neuropathy, and was also associated with increased severity of retinopathy and nephropathy. Another research confirmed that insulin and C-peptide deficiencies could cause the additional defects in protein kinase C activity, further resulting in severe Na⁺/K⁺-ATPase defect in type 1 diabetic peripheral neuropathy.³⁵ Besides, insulin and C-peptide deficiencies can also magnify impaired endothelial nitric oxide synthase (eNOS) expression and NO activity, abnormalities which might cause endoneurial hypoxemia in diabetic nephropathy.^{36,37}

Apart from providing structural support for proinsulin cleavage, C-peptide also has physiological functions on its own. C-peptide exhibits specific binding to cell surface with high affinity.³⁸ Although no specific C-peptide receptor has been identified yet, existing evidence indicates that C-peptide signals *via* the insulin signaling pathways.³⁹⁻⁴¹ By activating insulin receptor, C-peptide increases amino acid uptake and glycogen synthesis on its own. What's more, C-peptide shows a regulatory effect on insulin signaling, which is suppressive at high insulin levels and enhance at low insulin levels.⁴²

Protective functions of C-peptide in diabetes

Based on those findings, several studies have been conducted on the beneficial effects of C-peptide in diabetic cerebral disease. C-peptide treatment for 6 months was found improve sensory nerve function.^{43,44} Furthermore, C-peptide has been shown to prevent diabetic neuropathy in type 1 diabetes.⁴⁵ It exerts the effect by improving blood flow, preventing neuronal apoptosis, and suppressing diabetes-induced abnormal renal eNOS expression.^{46,47} In the vascular system, C-peptide has been shown to prevent vascular dysfunction in diabetic rats, and exert anti-proliferative effects on vascular smooth muscle cells, which may prevent atherosclerosis.^{32,48,49} The study group of Li and Sima has conducted some research on C-peptide in type 1 diabetes and found that administration of C-peptide prevents the neuronal apoptosis in hippocampus and partially corrects perturbed insulin-like growth factor activity,¹⁴ suggesting a relationship between C-peptide

deficiency and neuronal apoptosis.

In summary, former researches and reviews indicate that insulin/C-peptide deficiency play important roles in the occurrence and development of diabetic encephalopathy.

RISK FACTOR CONTROL AND THERAPIES FOR DIABETIC ENCEPHALOPATHY

Diabetic encephalopathy contributes to the morbidity and mortality associated with diabetes, thus partially bringing heavy economic burden onto the health care system. Finding effective measures to control the risk factors of this disease and new potential therapies would be conducive to addressing this problem. As described above, the most significant risk factors are hyperglycemia and abnormal C-peptide level. It has been suggested that improvements in glycemic control and C-peptide level can reduce the risk of diabetic cerebral disease.⁵⁰ Exogenous C-peptide administration has beneficial effects in many tissues commonly affected by diabetic complications including encephalopathy. In addition, C-peptide improves diabetes-affected erythrocyte deformability, thus improving oxygen availability and uptake in affected tissues. Increasing studies report a reduction of micro- and macrovascular complications in patients with type 1, as well as type 2, diabetes when the circulating concentration of C-peptide is close to physiological level.³² Pancreatic transplantation, which restores not only insulin secretion but also secretion of C-peptide, is associated with prevention and even reversal of diabetic complications including diabetic cerebral disease.⁵¹ Clearly, better control of these two risk factors in diabetic patients can bring more favorable outcomes and prevent diabetic encephalopathy.

Over the past decades, strategies aiming at reducing diabetic complications have focused not only on optimizing the metabolic control and normalizing blood glucose levels but also on the reduction/blockade of hyperglycemia-induced pathomechanisms. Protection from diabetes-induced dysfunction by C-peptide administration has been reported in clinical practice,⁵² yet given the small scale of that clinical trial, long-term large-scale studies are still needed for determination of the safety of long-term C-peptide administration in diabetic patients. It is not unlikely that C-peptide substitution in physiological doses would prevent endothelial dysfunction and reduce the risk of diabetes complications.³² Generally speaking, an early intensified antidiabetic treatment is critically significant for the prevention of diabetic complications, especially the prevention of diabetic cerebral disease.

CONCLUSIONS

Diabetic encephalopathy is characterized by neurophysiological impairments, structural changes, and cognitive dysfunction.⁵³ The underlying mechanisms of this disease are not fully illuminated yet. Clinical researches may attract some attention on the early cerebral functional changes and the prognosis of this disease.

Insulin/C-peptide deficiency in diabetes may be one of the most important pathogenic factors of diabetic encephalopathy. The mechanisms involving C-peptide, that are thought to be closely related to diabetic encephalopathy, present ample opportunity for further studies and for potential targets in therapeutic trials.

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