Antidiabetic Actions of Arachidonic Acid and Zinc in Genetically Diabetic Goto-Kakizaki Rats

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In previous studies, we showed that feeding arachidonic acid (AA) supplemented with a fixed amount of zinc lowered blood glucose concentrations in the fed state and water intake in rats with streptozotocin-induced diabetes. The present study was designed to determine dose-dependent effects of AA supplemented with a fixed amount of zinc on fed blood glucose levels, water intake, and glucose tolerance in genetically type 2 diabetic Goto-Kakizaki (G-K) Wistar rats. In an acute study, 20 mg/kg AA plus 10 mg/kg zinc administered via gastric gavage significantly improved oral glucose tolerance in G-K rats when compared to rats given distilled water (DW) only. When rats were treated chronically (2 weeks) with increasing doses of AA in drinking water, fed blood glucose concentrations and water intake were maximally decreased with diets containing 20 or 30 mg/L AA plus 10 mg/L zinc. Three-hour average area-above-fasting glucose concentrations (TAFGC; index of oral glucose tolerance) in diabetic G-K rats treated with 10, 20, or 30 mg/L AA plus 10 mg/L zinc for 2 weeks were significantly decreased relative to DW-treated rats. The effect on TAFGC values was maintained for an additional 2 weeks after cessation of treatment. Plasma insulin levels significantly increased in rats treated with 20 mg/L AA only or 10 mg/L AA plus 10 mg/L zinc, but not in rats treated with 20 or 30 mg/L AA plus 10 mg/L zinc, which are the most effective doses for the improvement of clinical signs of diabetes in G-K rats. In in vitro assays, 0.2 mg/mL AA in the incubation media was optimal for glucose uptake in isolated soleus muscle slices. These results suggest that treatment of genetically diabetic G-K rats with AA plus zinc lowers blood glucose levels via improvement of insulin sensitivity.

WE HAVE PREVIOUSLY demonstrated that arachidonic acid (AA) supplemented with zinc (ZA) improved clinical signs of diabetes in rats treated with streptozotocin.[1,2] This animal model is relevant to type 1 diabetes in humans. Type 1 diabetes is a genetic disease of pancreatic β-cell destruction evoked by certain environmental factors. Although AA stimulates insulin secretion in isolated pancreatic β cells,[3] the effectiveness of oral ZA treatment in these rats suggests that ZA possibly enhances glucose utilization, as no insulin secreatagogue can stimulate insulin secretion in pancreatic β cells damaged by streptozotocin treatment. Thus, we hypothesized that if ZA improves glucose tolerance in type 2 diabetic animals, the ZA effect that results in decreased blood glucose levels must be obtained through enhanced insulin sensitivity. To test this hypothesis, the effects of ZA on the improvement of clinical signs of diabetes were determined in genetically type 2 diabetic Goto-Kakizaki (G-K) rats.

The G-K rat strain was developed in Japan through inbreeding of mutant Wistar rats, and is a polygenic model of human type 2 diabetes. Type 2 diabetes or non–insulin-dependent diabetes mellitus (NIDDM) involves a syndrome of insulin resistance, including resistance to insulin-mediated signal transduction mechanisms.[4] Therefore, determination of the effects of ZA on the improvement of clinical signs of type 2 diabetes in G-K rats is important prior to the elucidation of the mechanisms by which ZA improves hyperglycemia. Zinc is involved in the stimulation of insulin-receptor signal transduction mechanisms.[5,6] Treatment of diabetes with zinc alone does not improve clinical conditions of diabetes[1,2,7,8]; however, it is possible that increased AA intake may control blood glucose level by regulating zinc metabolism.[9] The present study demonstrates that dietary intake of ZA improves clinical signs of type 2 diabetes in genetically diabetic G-K rats.

MATERALS AND METHODS

Materials

Zinc, AA, and L-histidine (a zinc-chelating agent) were purchased from Sigma Chemical Co (St Louis, MO). Accu-Chek Glucometer and strips were purchased from Roche Diagnostics Corp (Indianapolis, IN). The enzyme-linked immunosorbent assay (ELISA) kit specific for rat insulin was supplied by American Laboratory Products Co (Windham, NH). The compounds 1, 2-^{3}H-2-deoxy-D-glucose and 1-^{14}C-D-mannitol were obtained from Perkin Elmer Life Sciences (Boston, MA). Abbot Laboratories (North Chicago, IL) supplied pentobarbital.

Animals

Two-month-old Wistar rats were purchased from Charles River Laboratories, Indianapolis, IN. One-month-old male and female stock of G-K rats were purchased from the University of South Florida, Division of Comparative Medicine (Dr Robert J. Farese) and the colony was maintained at the Animal Facility of the Veterans Affairs (VA)

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Submitted March 14, 2001; accepted September 5, 2002.

Supported in part by Grant No. 1 R43 DK 56546-01 from the National Institutes of Health (to M.K.S.).

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0026-0495/03/5201-0040$35.00/0

doi:10.1053/meta.2003.50031

Greater Los Angeles Healthcare System, Los Angeles, CA. These studies were conducted with the approval of the Animal Study Committee at the VA Greater Los Angeles Healthcare System.

Experimental Methods and Design

Measurements of blood glucose and plasma insulin. Blood glucose was measured using an Accu-Chek Glucometer. One drop of blood was obtained from a cut on the underside of the rat's tail. At the end of the feeding experiment, rats were anesthetized by intraperitoneal injection of 100 mg/kg pentobarbital and heparin to prevent blood coagulation. Rats were exsanguinated via ocular vein puncture and blood was centrifuged at 1,000 × *g* for 5 minutes to collect plasma. Plasma insulin levels were measured by using an ELISA kit specific for rat insulin.

Measurement of 3-hour average area-above-fasting glucose concentrations (TAFGC). TAFGC, indices of glycemic control, were measured by determining blood glucose levels every 30 minutes for 3 hours after gastric gavage with 1.0 g glucose/kg body weight (BW). TAFGC were calculated by subtracting fasting blood glucose from the average blood glucose calculated during the 3 hours after glucose gavage.

Effects of ZA on muscle glucose uptake. Hind limb muscle tissues were removed from 2-month old nondiabetic Wistar and diabetic G-K rats after they were killed by 100 mg/kg BW pentobarbital injection. Tissue was sliced finely with a razor blade in Krebs Henseleit Buffer (KHB) solution. KHB Stock Solution (10X) consists of 118.5 mmol/L NaCl, 4.7 mmol/L KCl, 3.4 mmol/L $CaCl_2$, 1.2 mmol/L KH_2PO_4, 1.2 mol/L $MgSO_4$, 25 mmol/L $NHCO_3$, 32 mmol/L mannitol, 8 mmol/L glucose, and 0.1 % wt/vol bovine serum albumin (BSA). Slices were incubated in 15-mL polypropylene test tubes containing 2.0 mL Eagle's media with increasing levels of AA (0, 0.2, 0.5, 1.0, or 3.0 mg/mL), and 1.5 μCi 1,2-^{3}H-2-deoxy-D-glucose (specific activity, 1.0 μCi/mg) plus 0.3 μCi 1-^{14}C-D-mannitol (specific activity, 0.1 μCi/mg). Tubes were incubated at 35°C for 20 minutes. At the end of the incubation period, tissue samples were washed twice with KHB solution, dried in a 90°C oven overnight, weighed, reconstituted in distilled water, and frozen. Each tissue sample was homogenized in a Polytron homogenizer, centrifuged for 5 minutes at 1,000 × *g,* and the supernatant placed in a 20-mL vial for ^{3}H and ^{14}C counting. The precipitate was washed with 2 mL KHB solution, and added to the vial containing 2.0 mL initial supernatant. After addition of 15 mL liquid scintillation cocktail, the vials were counted in a Beckman liquid scintillation counter (Palo Alto, CA). The total amount of glucose bound and transported into tissue was determined by the measurement of radioactivity of 1, 2-^{3}H-2-deoxy-D-glucose. The amount of glucose bound to the tissue but not transported was determined by measuring radioactivity of 1-^{14}C-D-mannitol. Total tissue glucose uptake was calculated by subtracting tissue-bound glucose from total glucose bound to the cell membranes and transported into the cells.

Experimental Design

Determination of acute effects of ZA on TAFGC. For determination of acute effects of ZA on TAFGC, food was withdrawn for 4 hours in 2-month-old diabetic G-K rats. Rats were then administered with DW only, 10 mg/kg BW zinc only, 20 mg/kg BW AA only, or 10 mg/kg zinc plus 10, 20, or 30 mg AA/kg BW together with 1.0 g glucose/kg BW via gastric gavage (n = 5 to 10 for each treatment group). Blood glucose levels were measured every 30 minutes for 3 hours.

Determination of long-term effects of ZA on the clinical signs of diabetes. To measure the optimal concentration of AA required to lower blood glucose after long-term treatment, 1-month-old male G-K rats were divided into 6 groups of 5 to 10 rats. Rats were treated with either DW only, 10 mg/L zinc only, 20 mg/L AA only, or increasing levels of AA (10, 20, or 30 mg/L) plus 1.0 mg zinc and 0.5 mg L-histidine/L for 2 weeks. Every other day for 2 weeks, fed blood glucose and water intakes were measured at the same time in the morning. TAFGC were determined at the end of the 2-week treatment period and 2 weeks after cessation of treatments. Food was withdrawn overnight and animals were given 1.0 g/kg BW glucose via gastric gavage to determine the TAFGC. Feeding plasma insulin levels were determined in fed animals 2 weeks after cessation of treatments.

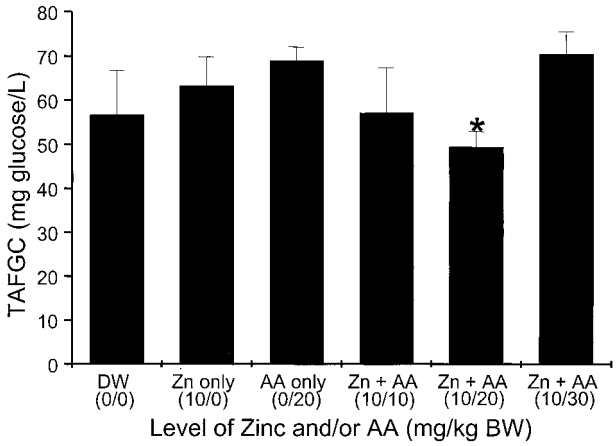

Fig 1. Acute effects of AA plus zinc on TAFGC in diabetic G-K rats after gastric gavage of glucose. Blood glucose levels were measured every 30 minutes for 3 hours after gastric gavage of 1 g glucose/kg BW with DW only, 10 mg/kg BW zinc, 20 mg/kg BW AA, or 10 mg zinc, and 0.5 mg L-histidine/kg BW plus 10, 20, or 30 mg AA/kg BW in 2 month-old G-K rats (n = 5-10 for each). *P < .05 compared to 20 mg/kg AA only-treated rats.

Statistical Analysis

Values are presented as the mean ± SEM. Data were analyzed by repeated-measures analysis of variance (ANOVA; blood glucose) or *t* test using a statistical package, GraphPad InStat (Version 1.13, supplied by GraphPad Software Co, San Diego, CA). A P value less than .05 was considered statistically significant.

RESULTS

For determination of acute effects of ZA on glucose disposal in genetically diabetic G-K rats, TAFGC were determined. When the 4-hour fasted G-K rats were administered DW only, 10 mg/kg BW zinc only, 20 mg/kg BW AA only, or 10 mg zinc/kg BW plus increasing doses of AA (10, 20, or 30 mg AA/kg BW), the maximal decrease in TAFGC was observed in the rats treated with 20 mg AA plus 10 mg zinc/kg BW (Fig 1). The TAFGC values in these rats were significant compared to the values in rats treated with 20 mg/kg BW AA only (P < .05).

For determination of the long-term effects of ZA on improvement of clinical signs of diabetes, G-K rats were treated with DW, 10 mg/L zinc only, 20 mg/L AA only, or 10 mg/L zinc plus 10, 20 or 30 mg/L AA for 2 weeks. Fed blood glucose levels at day 14 were significantly lower in G-K rats treated with drinking water containing 20 or 30 mg AA/L plus 10 mg/L zinc compared to those treated with DW only (Fig 2). Rats treated with 10 mg/L zinc plus 10 or 30 mg/L AA showed similar effects as rats given 20 mg/L AA plus 10 mg/L zinc. To determine TAFGC, food was withheld overnight and animals

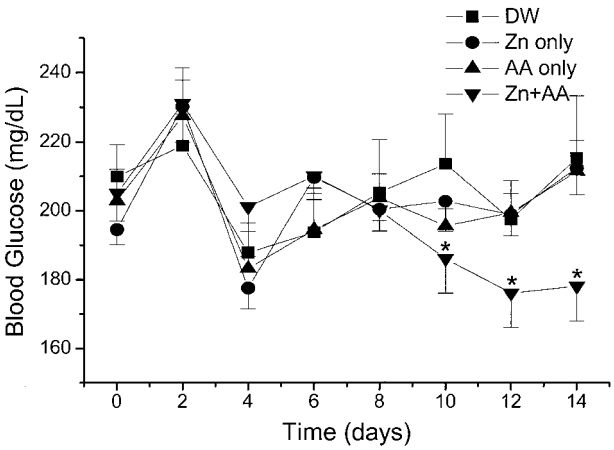

Fig 2. Blood glucose levels in fed G-K rats treated with various doses of zinc and AA. One-month-old diabetic G-K rats were treated with DW only, 10 mg/L zinc only, 20 mg/L AA only, or 10 mg/L zinc plus 10, 20, or 30 mg/L AA in drinking water (n = 5-10) for 2 weeks and blood glucose levels measured every other day. The 10 mg/L and 30 mg/L AA plus 10 mg/L zinc-treated rats showed the similar values to those of 20 mg/L AA plus 10 mg/L zinc; therefore, data for these groups are not shown. *$P < .05$ compared to DW-treated rats.

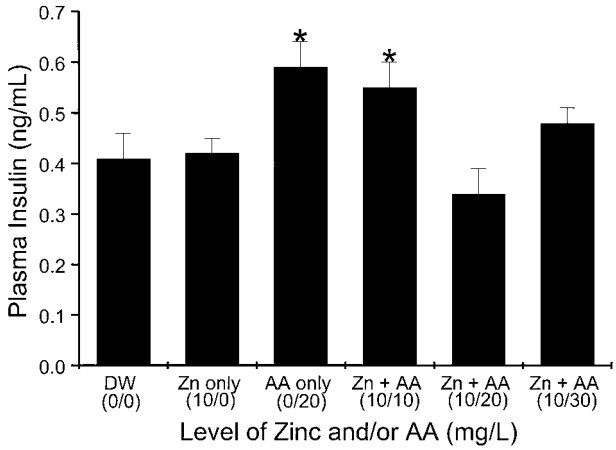

Fig 4. Plasma insulin levels in G-K rats treated with various doses of zinc and AA. Fed rats were treated with DW only, 10 mg/L zinc only, 20 mg/L AA only, or 10 mg/L zinc plus 10, 20, or 30 mg/L AA in drinking water for 2 weeks (n = 5-10). Plasma insulin levels were determined 2 weeks after cessation of treatment. *$P < .05$ compared to DW-only treatment group.

were given 1.0 g/kg glucose via gastric gavage. TAFGC values in G-K rats treated with 10 mg/L zinc only or 20 mg/L AA only for 2 weeks did not show any significant difference compared to those treated with DW. However, all rat groups treated with increasing doses of AA (10, 20, or 30 mg/L) plus 10 mg/L zinc showed significantly improved TAFGC values compared to DW-treated rats (Fig 3). When these rats were monitored for another 2 weeks, those previously treated with 10, 20, or 30 mg/L AA plus 10 mg/L zinc still maintained significantly improved TAFGC values compared to DW-treated rats.

Plasma insulin levels increased significantly in the rats treated with 20 mg/L AA only or 10 mg/L zinc plus 10 mg/L AA compared to those treated with DW or zinc only ($P < .05$) (Fig 4). However, no significant changes were observed with 20 or 30 mg/L AA plus 10 mg/L zinc-treated rats compared to DW-treated rats. Water intake was decreased most in rats treated with either 20 or 30 mg/L AA plus 10 mg/L zinc. The rats treated with either 10 mg/L zinc only or 10 mg/L AA plus 10 mg/L zinc also significantly decreased water intake (Fig 5).

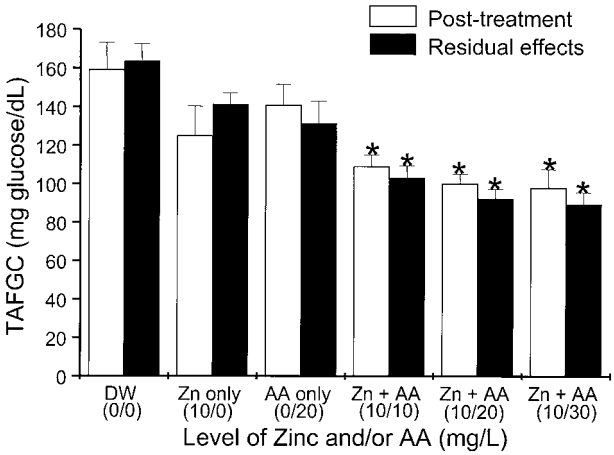

Fig 3. Values of TAFGC in G-K rats treated with various doses of zinc and AA for 2 weeks (Post-treatment) and measured 2 weeks after cessation of treatment (Residual effects). G-K rats were divided into 6 groups of 5-10 rats, and treated with DW only, 10 mg/L zinc only, 20 mg/L AA only, or 10 mg/L zinc plus 10, 20, or 30 mg/L AA in drinking water for 2 weeks. At the end of the treatment period, rats were fasted overnight and blood glucose levels were measured every 30 minutes after gavage of 1 g/kg BW glucose (n=5-10 for each) (□). Two weeks after cessation of treatment, TAFGC values were measured again (■). * $P < .05$ compared to DW-treated rats.

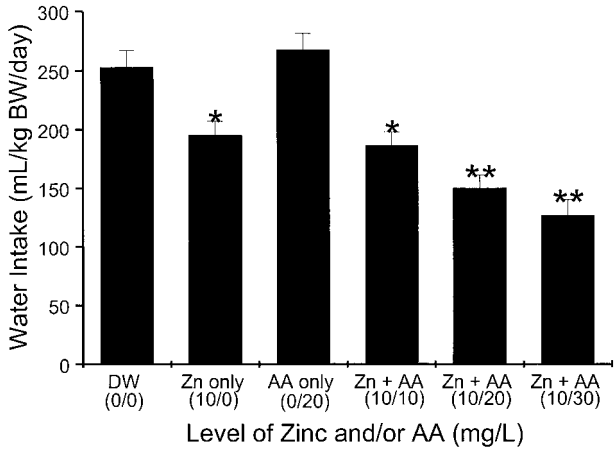

Fig 5. Water intake of G-K rats treated with various doses of AA plus zinc in drinking water. Diabetic G-K rats were treated with DW only, 10 mg/L zinc only, 20 mg/L AA only, or 10 mg/L zinc plus 10, 20, or 30 mg/L AA in drinking water for 2 weeks and water intake was measured every other day (n = 5-10). **$P < .01$ compared to DW-only treatment group.

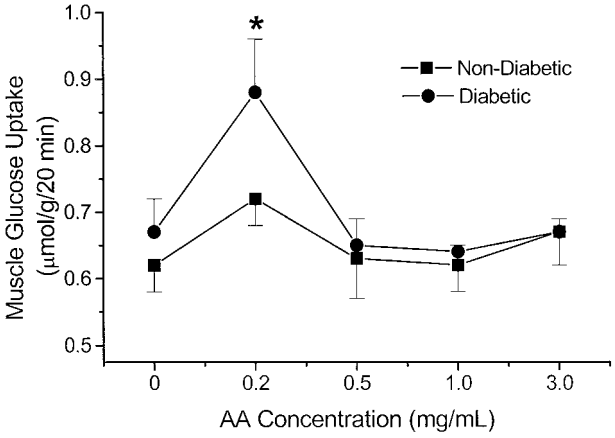

Fig 6. AA concentration-dependent glucose uptake capacities of Wister nondiabetic and G-K rat muscle tissues. Muscle tissues (soleus) from age-matched Wistar nondiabetic and diabetic G-K rats (n = 10 for each) were removed and incubated in Eagle's media with increasing levels of AA (0, 0.2, 0.5, 1.0, or 3.0 mg/mL), and 1.5 μCi 1, 2-^{3}H-2-deoxy-D-glucose plus 0.3 μCi 1-^{14}C-D-mannitol at 35°C for 20 minutes. *P < .05 compared to 0 mg/mL AA concentration.

Thus, water intake was reduced in rats with all treatments containing zinc relative to DW-treated rats, but not with the AA-only treatment.

When effects of ZA on glucose uptake rates in muscle tissues from normal Wistar and diabetic G-K rats were determined, the optimal concentration of AA for glucose uptake in muscle slices was 0.2 mg/mL for both normal Wistar and diabetic G-K rats (Fig 6). At the optimal concentration of AA, glucose uptake capacity of muscle slices from G-K rats was higher than that from normal rats.

DISCUSSION

AA is the major prostaglandin (PG) precursor. Although the mechanisms are not clearly understood, AA and PG play an important role in the regulation of insulin release and participate in numerous glucose metabolic activities.[10-12] PGE_2 and PGE_1 treatment improves insulin sensitivity in isolated rat soleus muscle[13] and confers therapeutic benefit to diabetic patients.[14,15] Blood levels of AA are decreased in diabetic rats,[16] and lack of free fatty acids in muscle tissues is associated with increased insulin resistance in humans.[17] Although AA stimulates insulin synthesis and secretion,[3] it has been suggested that supplementation of γ-linoleic acid (GLA), which is a precursor of AA, may diminish some of the complications of type 2 diabetes by increasing AA synthesis.[18] PGs and essential fatty acids are capable of chelating zinc and regulating intestinal zinc absorption and secretion.[19-22] AA influences zinc uptake capacity (from mucosa to serosa) in intestinal segments and in various organ tissues,[22,23] and regulates muscle zinc metabolism.[9] These facts illustrating that PGs and AA affect both glucose[1-3,10-13] and zinc[19-22] metabolism, and that zinc is an important stimulator of insulin-mediated signal transduction mechanisms leading to increased glucose uptake in insulin-requiring cells,[5,6] suggest that AA influences zinc metabolism, which may in turn improve clinical signs of diabetes.

We found in this study that the maximal improvement of TAFGC values occurred when rats were administered acutely 20 mg AA/kg BW via gastric gavage (Fig 1). The optimal dose of AA for glucose uptake rate in isolated muscle tissue was 0.2 mg/mL in the cell culture medium (Fig 6). As approximately 5% of total body weight is plasma, a 1.0 kg rat would have a plasma volume of 50 mL. Thus, 20 mg AA/50 mL plasma volume is equivalent to 0.4 mg/mL. Assuming only 50% of total AA administered is absorbed, the blood concentration attainable with the optimal oral dose of AA (Fig 1) in the in vivo studies is remarkably similar to the optimal AA concentration in the muscle tissue incubation found in the in vitro studies (Fig 6). Given that AA nutrition in diabetic rats is defective and that the AA requirement in these animals may be different from that of normal rats, it is apparent that AA supplementation can improve the clinical conditions of diabetes. However, it is important to maintain adequate but not excessive AA intake. Since AA can be a PG precursor and competitor of PG binding sites, AA may compete with PG activities if excess AA is present in the plasma. Additionally, AA can stimulate zinc metabolism by enhancing PG synthesis, which in turn improves insulin secretion or glucose utilization of muscle tissues.[1-3,10-13,19-22] It is possible that excess AA may interfere with PG action by acting as a PG analog. Further studies are required to fully determine the dose-dependent mechanisms of AA's effects to lower blood glucose levels.

The effect of acute administration of ZA (20 mg/kg BW AA) in lowering TAFGC, an index of oral glucose tolerance, could be due to the increased plasma insulin levels or direct (non–insulin-dependent) effects on muscle tissues to improve insulin action. It has been reported that both AA[3] and zinc[23,24] stimulate insulin secretion, and glucose uptake in muscle tissues.[9] However, improvement of TAFGC took an extended length of time in the long-term studies using low doses of AA, and the effect was sustained even after cessation of treatment. The major defect of type 2 diabetes is impaired insulin action to enhance glucose uptake due to impaired cellular signal transduction mechanisms. AA is an essential element to maintain integrity of cell membranes; replenishment of AA from the deficient state may act to correct the chemical structure of cell membranes and reverse impaired signal transduction. Furthermore, AA is the major PG precursor for the control of cellular function, and contributes to cellular PG synthesis. Zinc is an extremely important essential element, since it is involved in gene expression and metabolism of cellular waste,[25] and acts as a cofactor for insulin receptor β-subunit autophosphorylation.[5,6] Repair of cellular function by AA and zinc to improve glucose utilization by muscle and fat cells may take some time. Once cellular function is corrected, the treatment effect may last without further treatment as shown in Fig 3. Further work is needed to elucidate the mechanism for this sustained effect.

ZA may have several actions to improve diabetes, including stimulating insulin secretion,[10-12] and/or direct (non–insulin-dependent) glucose uptake in muscle tissues[5,6] that results in an immediate effect in lowering blood glucose levels. ZA may improve insulin action by stimulating glucose utilization in

muscle tissues, which requires more time for metabolic adjustment for cellular function and structure.[10-13,18] This possibility is further supported by the data showing that rats treated with 20 or 30 mg/L AA did not show any increase in plasma insulin levels (Fig 4), although these rats demonstrated improved TAFGC values (Fig 3). However, AA treatment increased plasma insulin levels in 20 mg/L AA only-treated rats concurrently with no improvement in glucose utilization (Figs 3 and 4). One of the major clinical conditions of diabetes is polydypsia. If diabetic signs are improved by ZA treatment, it is expected that water intake in rats treated with ZA must decrease. As shown in Fig 5, water intake rate did decrease with 20 or 30 mg/L AA plus 10 mg/L zinc treatments.

Insulin resistance is present 10 to 20 years before the onset of type 2 diabetes,[26] and epidemiological studies confirm the incidence of type 2 diabetes is directly related to pre-existing insulin resistance.[27,28] A major biochemical defect in insulin resistance is impaired action of the insulin receptor γ-subunit autophosphorylation.[29-32] This defect is improved by zinc, which enhances insulin action[10-13,18] and even has insulin-like properties in itself.[5,6] AA is capable of chelating zinc and stimulating intestinal tissue zinc uptake and absorption.[33,34] Thus, AA plus zinc can improve glucose tolerance by increasing glucose utilization in muscle tissues.

CONCLUSIONS

Although the pathophysiology of type 2 diabetes in the G-K rat model may not be exactly identical to that of the human, the improvement demonstrated in the G-K rats' clinical signs of diabetes suggest the possibility of treating human diabetes with AA plus zinc. However, it is difficult to assess the treatment value of short-term treatment with AA plus zinc on G-K rats. The fact that G-K rats maintained improved TAFGC values for a few weeks after the cessation of treatment implies that the long-term treatment of diabetic G-K rats with various concentration of AA plus zinc may have as yet unknown treatment value. Nevertheless, evidence of improvement in diabetes in G-K rats with AA plus zinc treatment is presented in this study, although the exact mechanisms have yet to be fully determined.

ACKNOWLEDGMENT

We are grateful to Anita Stein and Debra Wong for their editorial assistance. We also would like to thank Dr Suk J. Hong for his assistance in determining muscle glucose uptake and Dr Yong Liu for his assistance in determining plasma insulin.

REFERENCES

1. Song MK, Rosenthal MJ, Kang KW, et al: Animal prostate extract ameliorates diabetic symptoms by stimulating intestinal zinc absorption in rats. Diabetes Res 31:157-170, 1996

2. Song MK, Rosenthal MJ, Hong SJ, et al: Synergistic antidiabetic activities of zinc, cyclo (his-pro), and arachidonic acid. Metabolism 50:53-59, 2001

3. Miller GD, Keen CL, Stern JS, et al: Copper deficiency and arachidonic acid enhance insulin secretion in isolated pancreatic islets from lean (FaFa) Zucker rats. Pancreas 17:390-396,1998

4. Reaven GM: Pathophysiology of insulin resistance in human disease. Physiol Rev 75:473-486,1995

5. Ezaki O: IIb group metal ions (Zn^{2+}, Cd^{2+}, Hg^{2+}) stimulate glucose transport activity by post-insulin receptor kinase mechanism in rat adipocytes. J Biol Chem 264:16118-16122, 1989

6. Tang X, Shay NF: Zinc has an insulin-like effect on glucose transport mediated by phosphoinositol-3-kinase and Akt in 3T3-L1 fibroblasts and adipocytes. J Nutr 131:1414-1420, 2001

7. Brandao-Neto J, da Silva CA, Figueiredo NB, et al: Lack of acute zinc effects in glucose metabolism in health and insulin-dependent diabetes mellitus patients. Biometals 12:161-165, 1999

8. Blostein-Fujii A, DiSilvestro RA, Frid D, et al: Short-term zinc supplementation in women with non-insulin-dependent diabetes mellitus: Effects on plasma 5′-nucleotidase activities, insulin-like growth factor I concentrations, and lipoprotein oxidation rates in vitro. Am J Clin Nutr 66:639-642, 1997

9. Rosenthal MJ, Hwang IK, Song MK: Effects of arachidonic acid and cyclo (his-pro) on zinc transport across small intestine and muscle tissues. Life Sci 70:337-348, 2001

10. Aalusha PV, Caldwell A: Prostaglandins and diabetes mellitus, in Ellenberg M, Rifkin H (eds): Diabetes Mellitus. Theory and Practice (ed 3). New York, NY, Medical Examination Publishing Co, 1983, pp 295-308

11. Robertson RP: Prostaglandins, glucose homeostasis and diabetes mellitus. Med Clin North Am 65:759-771, 1984

12. Robertson RP, Chen M: A role for prostaglandin E in defective insulin secretion and carbohydrate intolerance in diabetes mellitus. J Clin Invest 60: 747-753, 1977

13. Leighton B, Budohoski L, Lozeman F, et al: The effect of prostaglandins E1, E2 and F2α and indomethacin on the sensitivity of glycolysis and glycogen synthesis to insulin in the stripped soleus muscle of the rat. Biochem J 227:337-340, 1985

14. Abe K, Ozono Y, Miyazaki M, et al: Prostaglandin E1 for renal papillary necrosis in a patient with diabetes mellitus. J Int Med Res 27:90-95, 1999

15. Wentzel P, Eriksson UJ: Antioxidants diminish developmental damage induced by high glucose and cyclooxygenase inhibitors in rat embryos in vitro. Diabetes 47:677-684, 1998

16. Karageuzyan KG, Vartanyan GS, Agadjanov MI, et al: Restoration of the disordered glucose-fatty acid cycle in alloxan-diabetic rats by trihydroxyoctadecadienoic acids from Bryonia alba, a native Armenian medicinal plant. Planta Med 64:417-422, 1998

17. Clore JN, Harris PA, Li J, et al: Changes in phosphatidylcholine fatty acid composition are associated with altered skeletal muscle insulin responsiveness in normal man. Metabolism 49:232-238, 2000

18. Jamal G: Prevention and treatment of diabetic distal polyuneuropathy by the supplementation of γ-linoleic acid, in Horrobin DF (ed): Omega-6 Essential Fatty Acids: Pathophysiology and Roles in Clinical Medicine. New York, NY, Wiley-Liss, 1990, pp 485-504

19. Song MK, Adham NF: Role of prostaglandin E2 in zinc absorption in the rat. Am J Physiol 234:E99-105, 1978

20. Bukhave K, Rask-Madsen J: Saturation kinetics applied to in vitro effects of low-prostaglandin E2 and F2α concentrations on ion transport across human jejunal mucosa. Gastroenterology 78:32-42, 1980

21. Song MK, Lee DB, Adham NF: Influence of prostaglandins on unidirectional zinc fluxes across the small intestine of the rat. Br J Nutr 59:417-428, 1988

22. Song MK, Adham NF: Evidence for an important role of pros-

taglandins E2 and F2 in the regulation of zinc transport in the rat. J Nutr 109:2152-2159, 1979

23. Huber AM, Gershoff SN: Effect of zinc deficiency in rats on insulin release from the pancreas. J Nutr 103:1379-1744, 1973

24. Chausmer AB: Zinc, insulin and diabetes. J Am Coll Nutr 17:109-115, 1998

25. Eichhorn GL: Aging, genetics, and the environment: potential of errors introduced into genetic information transfer by metal ions. Mech Ageing Dev 9:291-301, 1979

26. Warram JH, Martin BC, Krolewski AS, et al: Slow glucose removal rate and hyperinsulinemia precede the development of type II diabetes in the offspring of diabetic parents. Ann Intern Med 113:909-915, 1990

27. Reaven GM, Bernstein R, Davis B, et al: Nonketotic diabetes mellitus: Insulin deficiency or insulin resistance? Am J Med 60:80-88, 1976

28. Defronzo RA: Lilly Lecture 1987. The triumvariate: Beta-cell, muscle, liver. A collusion responsible for NIDDM. Diabetes 37:667-687, 1988

29. Amer P, Pollare T, Lithell H, et al: Defective insulin receptor tyrosine kinase in human skeletal muscles in obesity and type 2 (non-insulin-dependent) diabetes mellitus. Diabetologia 30:437-440, 1987

30. Caro JF, Sinha MK, Raju SM, et al: Insulin receptor kinase in human skeletal muscle from obese subjects with and without noninsulin dependent diabetes. J Clin Invest 79:1330-1337, 1987

31. Maegawa H, Shigeta Y, Egawa K, et al: Impaired autophosphorylation of insulin receptors from abnormal skeletal muscles in nonobese subjects with NIDDM. Diabetes 40:815-819, 1991

32. Obermaier-Kusser B, White MF, Pongratz DE, et al: A defective intramolecular autoactivation cascade may cause the reduced kinase activity of the skeletal muscle insulin receptor from patients with non-insulin-dependent diabetes mellitus. J Biol Chem 264:9497-9504, 1989

33. Song MK, Mooradian AD: Intestinal zinc transport: Influence of streptozotocin-induced diabetes, insulin and arachidonic acid. Life Sci 42:687-694, 1988

34. Song MK, Littner MR, Adham NF, et al: Effect of oral administration of arachidonic acid on prostaglandin and zinc metabolism in plasma and small intestine of the rat. Prostaglandins Leukot Med 17:159-166, 1985