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Glycine treatment decreases proinflammatory cytokines and increases interferon-γ in patients with Type 2 diabetes

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ABSTRACT. ***Background:*** **Amino acids have been shown to stimulate insulin secretion and decrease glycated hemoglobin (A1C) in patients with Type 2 diabetes.** ***In vitro*****, glycine reduces tumor necrosis factor (TNF)-α secretion and increases interleukin-10 secretion in human monocytes stimulated with lipopolysaccharide. The aim of this study was to determine whether glycine modifies the proinflammatory profiles of patients with Type 2 diabetes.** ***Materials/subjects and methods:*** **Seventy-four patients, with Type 2 diabetes were enrolled in the study. The mean age was 58.5 yr, average age of diagnosis was 5 yr, the mean body mass index was 28.5 kg/m², the mean fasting glucose level was 175.5 mg/dl and the mean A1C level was 8%. They were allocated to one of two treatments, 5 g/d glycine or 5 g/d placebo, po tid, for 3 months.** ***Results:*** **A1C levels of patients given glycine were significantly lower after 3 months of treatment than those of the placebo group. A significant reduction in TNF-receptor I levels was observed in patients given glycine compared with placebo. There was a decrease of 38% in the interferon (IFN)-γ level of the group treated with placebo, whereas that of the group treated with glycine increased up to 43%. These data showed that patients treated with glycine had a significant decrease in A1C and in proinflammatory cytokines and also an important increase of IFN-γ.** ***Conclusion:*** **Treatment with glycine is likely to have a beneficial effect on innate and adaptive immune responses and may help prevent tissue damage caused by chronic inflammation in patients with Type 2 diabetes.**

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INTRODUCTION

The intake of amino acids has several effects on glucose metabolism. *In vitro* studies with pancreatic β cells have shown that arginine, leucine, phenylalanine, and glutamine have insulinotropic effects (1). However, some amino acids provoke undesirable effects; arginine is the best insulin secretor but induces diarrhea in animal models (1). Ingestion of glycine caused a modest reduction in the blood glucose of normal subjects and of patients with Type 2 diabetes (2). Furthermore, ingestion of glycine stimulated the insulin response to an oral glucose tolerance test (3). In the streptozotocin-induced diabetic rat model, glycine decreased blood glucose and hemoglobin 1C (A1C) levels and changed the lipid profile (4). A glycine-induced decrease in A1C levels was also reported by others, but the mechanism for A1C reduction is not known (5). Decreased glycation has been associated with modifications in the expression of interleukin (IL)-2 receptors on T cells and with alterations in the synthesis of IL-10 and IL-6. Such modifications may explain the low proliferative responses of T cells and peripheral blood mononuclear cells (PBMN) from patients with Type 2 diabetes and streptozotocin-induced diabetic rats to phytohemaglutinin (PHA) and other mitogens (6).

Moreover, glycine prevents cellular injury by a number of mechanisms, which include inhibition of the synthesis of proinflammatory cytokines, chemoattractants, and adhesion molecules; this effect has been observed in cells and organs after lipopolysaccharide (LPS) treatment, in ischemia-reperfusion of the liver, heart, and kidney, and in experimental models of organ transplants (7). Treatment of monocytes with glycine reduces the production of proinflammatory cytokines such as tumor necrosis factor (TNF)-α and increases the production of the anti-inflammatory cytokine IL-10 (7, 8).

The potential effects of glycine on cytokines and other inflammatory mediators is relevant to obesity and Type 2 diabetes because both conditions are associated with chronic, low-grade inflammation (9, 10). Several proinflammatory markers, such as the cytokine, IL-6 and the acute-phase C-reactive protein (CRP), as well as TNF-receptor I (RI) (a marker of TNF-α activity) are elevated in obese and insulin-resistant subjects (11-13). These markers are also elevated in subjects with impaired glucose tolerance or Type 2 diabetes (14, 15). The Atherosclerosis Risk in Communities Study demonstrated that markers of inflammation predict Type 2 diabetes (9). Those results were confirmed in the U.S. Women's Health Study as well as in the Pima Indians Study, in which CRP and IL-6 levels were elevated (16, 17).

Interferon (IFN)-γ is produced by activated T lymphocytes and natural killer (NK) cells also by NKT cells, and activates monocytes/macrophages and endothelial cells. IFN-γ stimulates the microbicidal activities of phagocytes, which is an advantage because it may help to eliminate

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microorganisms. There are few studies on the relationship between Type 2 diabetes and IFN-γ; in two studies it was observed that lymphocytes T (CD4+ and CD8+) from Type 2 diabetics released significantly lower amounts of IFN-γ in the intracellular space, compared with healthy volunteers (18, 19).
The aim of this study was to determine if glycine modifies the chronically proinflammatory state present in patients with Type 2 diabetes.

MATERIALS AND METHODS

Subject selection

Patients were recruited from Family Medicine Clinics of the Instituto Mexicano del Seguro Social (IMSS) in the metropolitan area of Mexico City. All subjects had previously been diagnosed with Type 2 diabetes in accordance with American Diabetes Association (ADA) criteria, and all participants had high blood glucose and A1C levels. Exclusion criteria were obesity [body mass index (BMI)>30 kg/m^2], heart disease or clinical evidence of complications of diabetes. All patients maintained their individual diets and oral anti-diabetic drug treatments during the trial. None received treatment with insulin. After selection by their family physicians, appointments were scheduled for the patients at the IMSS. Participants were evaluated a second time by a different physician after a 12-h fasting period. If the subjects still complied with the inclusion criteria they were asked to sign a consent form approved by the Comisión Nacional de Investigación Científica (Ethics Committee). Gender, age, previous treatments, and symptoms were recorded. Weight and height were measured using a clinical beam scale and a stadiometer. The patients wore light-weight clothing without shoes. BMI was expressed as kg/m^2. Circumferences of the waist (midway between the costal border and the iliac crest) and hip (between the great trochanters) were measured with a metric tape to the nearest 0.1 cm while the patient was standing. Blood pressure was measured once in a sitting position after 5 min of rest and twice at 5 min intervals thereafter. The first measurement was discarded, and the average of the second and third measurements was calculated. An antecubital vein blood sample for biochemical analysis was drawn after a 12 h fast. Patients were allocated randomly to one of two treatments (5 g glycine po tid or 5 g isocaloric placebo po tid) by means of a computer-generated list. The protocol was approved by the "Comisión Nacional de Investigación Científica" (Ethics Committee).

Treatment protocol

Glycine and placebo were supplied in envelopes as 5 g of white powder. The only identification mark on the enveloped consisted of a personal identification number. Patients were instructed to drink the powder dissolved in water tid (15 g/day). The optimal dose of glycine for decreasing the level of A1C was previously reported (20). Specialized personnel monitored that each person ingested glycine following the written instructions. Reagent grade glycine was purchased from Vitadrug SA de CV, México City. The placebo consisted of an isocaloric amount of starch (4 calories per g) (Vitadrug, SA de CV, México City). No further adjustments were made to the diet programs of the patients. Patients were scheduled to visit the clinic monthly after a 12 h fast. Any new symptoms or findings at their physical examination were recorded and the patients were given 1 month's supply of envelopes. The physician, patients, and other persons involved in the protocol did not know whether the envelopes contained glycine or placebo. A blood sample was drawn at the time of their last visit. In order to ascertain the effect of glycine in the AIC, we treated patients during a 3-month period.

Laboratory tests

All laboratory samples were taken from patients after an overnight 12 h fast. Aliquots of plasma and sera were immediately stored at –80 C. Biochemical determination of fasting glucose, total cholesterol, LDL-cholesterol (LDL-C), HDL-cholesterol (HDL-C), triglycerides, and A1C were performed automatically with a Synchron CX5 instrument (Beckman Instruments, Mexico City). Fasting insulin levels were assayed by chemiluminescence (Immulite, Mexico City). Insulin resistance (IR) was calculated using the homeostasis model assessment (HOMA) equation (21). All assays were performed in triplicate and expressed as means±SE.

Cytokines

IL-1β, 1L-6, TNF-RI, IFN-γ, and resistin were measured using commercial enzyme-linked immunosorbent assays (BD Bioscience Pharmingen®, San Diego, CA, and Peprotech®, Rocky Hill, NJ). Samples were analyzed in triplicate and expressed as means±SE.

Statistical analyses

All data were expressed as means±SE. All variables were contrasted with a two-way analysis of covariance; all comparisons were adjusted for sex, age, BMI, disease duration, and treatment (hypoglycemic agents, antihypertensives, etc). Non-normally distributed variables were first log-transformed. Correlations were evaluated using the Pearson correlation coefficient. All p-values <0.05 were considered significant. Statistical tests were performed using SPSS software (SPSS Inc., Chicago IL® v 10.0).

RESULTS

General characteristics of the subjects studied

Seventy-four patients were studied, of whom 40 (54%) were women. The average age was 58.5±10 yr and the average duration of the disease was 5.5±3.1 yr. Fifty-one (68.1%) patients were treated with hypoglycemic agents (80.5% in placebo and 57.8% in glycine groups respectively, p=0.035), 22 (29.7%) with antihypertensives, and 11 (14.9%) with hypolipemic agents. No patient was treated with thiazolidendiones. Mean (±SD) BMI was 28.8 (3.6) kg/m^2, mean waist circumference was 93.9 (9.1) cm, mean blood pressure was 120 (10)/ 81 (7) mmHg. Thirty-eight patients were allocated to the glycine treatment and 36 were allocated to the placebo treatment. There were no differences in baseline characteristics between groups (Table 1).

Effect of glycine treatment in patients with Type 2 diabetes

Results of 3 months of treatment are shown in Table 2. Fasting blood glucose levels were significantly lower after glycine treatment (183.3±57.6 *vs* 140.4±38.7) and placebo treatment (168.2±59.4 *vs* 150.4±44.0), differences between groups were nearly significant (p=0.0643). A1C levels were significantly lower after 3 months of ingestion of glycine (8.3±1.9 *vs* 6.9±1.3) or placebo

Table 1 - *Anthropomorphic and biochemical measurements of patients with Type 2 diabetes at the baseline study.*

Group No.	Placebo 36	Glycine 38
Age (yr)	59.5±9.6	57.5±9.8
Female sex (%)	21 (58%)	19 (50%)
Yr of diagnosis	5.3±2.3	5.4±3.7
Treatment no. (%):		
Diet	9 (25.0)	17 (44.0)
Hypoglycemic agents	29 (80.5) Glybenclamide 11 (30.5) Metformin 7 (19.4) Glybenclamide + Metformin 10 (27.7) Glybenclamide + Acarbose 1 (2.7)	22 (57.8) Glybenclamide 9 (23.7) Metformin 5 (13.1) Glybenclamide + Metformin 7 (18.4) Glybenclamide + Acarbose 1 (2.6)
Hypolipemic agents	3 (8.3) Bezafibrate 1 (2.7) Pravastatin 2 (5.5)	8 (21.0) Bezafibrate 3 (7.9) Pravastatin 5 (13.1)
BMI (kg/m²)	28.9±3.7	28.5±3.6
SBP (mmHg)	119.7±11	121.3±10.1
DBP (mmHg)	80.9±6.4	81.1±6.9
WHR	0.89±0.05	0.90±0.06
Fasting glucose (mg/dl)	168.2±59.4	183.3±57.6
A1C (%)	8.0±1.4	8.37±1.9

Values are means±SD; BMI: body mass index; SBP: systolic blood pressure; DBP: diastolic blood pressure; WHR: waist-hip ratio; A1C: A1C (%) fraction of glycated hemoglobin.

(8.0±1.4 *vs* 7.6±1.2); there was a greater reduction in the glycine group than in the placebo group (p=0.0364). There were no significant differences in mean fasting insulin levels between the beginning and end of glycine treatment (12.8±5.8 *vs* 15.2±4.9) or placebo treatment (13.6±7.4 *vs* 14.1±7.4) (p=0.5489). However, a significant reduction in HOMA-IR was observed (p=0.011) after 3 months, although there were no statistical differences between groups (glycine, 5.46±3.26 *vs* 4.95±2.35; placebo, 5.2±2.9 *vs* 5.1±2.7) (p=0.0867). No statistical differences were observed for the other variables shown in Table 2.

Proinflammatory profile after glycine ingestion

The effects of glycine ingestion on the proinflammatory profile are summarized in Figure 1. After 3 months of treatment, significant reductions in the levels of all cytokines were observed for both groups (glycine and placebo). However, for IL-1β, IL-6, and resistin, there were no statistically significant differences between groups (IL-1β, p=0.6688; IL-6, p=0.1486; resistin, p=0.0708). High baseline levels of TNF-RI were observed in both groups. After treatment, a significant reduction in TNF-RI levels was observed in patients given glycine treatment (1298.8±293.1 before treatment, 437.4±300.7 after treatment) compared with those given placebo (1355.2±230.7 before treatment, 829.18±419.59 after treatment) (p<0.0014). Interestingly, IFN-γ levels showed a different pattern, in that baseline levels were similar between groups, but after 3 months of treatment IFN-γ levels of the group given placebo decreased (463.09±133.72 *vs* 262.45±153.48) whereas those of the group treated with glycine increased significantly (448.76±156.84 *vs* 611.66 ±206.64) (p<0.0001).

Table 2 - *Effect of glycine after 3 months of treatment in patients with Type 2 diabetes.*

	Placebo basal	Placebo 3 months	Glycine basal	Glycine 3 months	*p*
Weight (kg)	72.1±10	72±10.1	72.4±12.2	71.9±12.1	0.948
BMI (kg/m²)	28.9±3.7	28.9±3.8	28.5±3.6	28.3±3.5	0.948
SBP (mmHg)	119.7±11	123.8±20.1	121.3±10.1	119.4±11.1	0.165
DBP (mmHg)	80.9±6.4	81.2±6.2	81.1±6.9	81.5±7.8	0.817
Fasting glucose (mg/dl)	168.2±59.4	150.4±44	183.3±57.6	140.4±38.7	0.0691
A1C (%)	8.0±1.4	7.6±1.2	8.3±1.9	6.9±1.3	0.0379
Total cholesterol (mg/dl)	202.9±36.4	205±50	196.7±33	200.6±41.5	0.869
LDL-C (mg/dl)	120.3±27.8	125.1±43.5	116.4±26	115±27	0.883
HDL-C (mg/dl)	44.6±12.5	41.9±12.3	42.5±12.5	40.4±19.9	0.900
Triglycerides (mg/dl)	198.2±90.9	188.7±90.7	238.4±129	219.4±115	0.684
Fasting insulin (μUI/ml)	13.6±7.7	14.1±7.4	12.8±5.8	15.2±4.9	0.584
HOMA-IR	5.2±2.9	5.1±2.7	5.46±3.26	4.95±2.53	0.0961

Values are means±SD. BMI: body mass index; SBP: systolic blood pressure; DBP: diastolic blood pressure; A1C: A1C (%) fraction of glycated hemoglobin. LDL-C: LDL cholesterol; HDL-C: HDL cholesterol; HOMA-IR: homeostasis model assessment-insulin resistance.

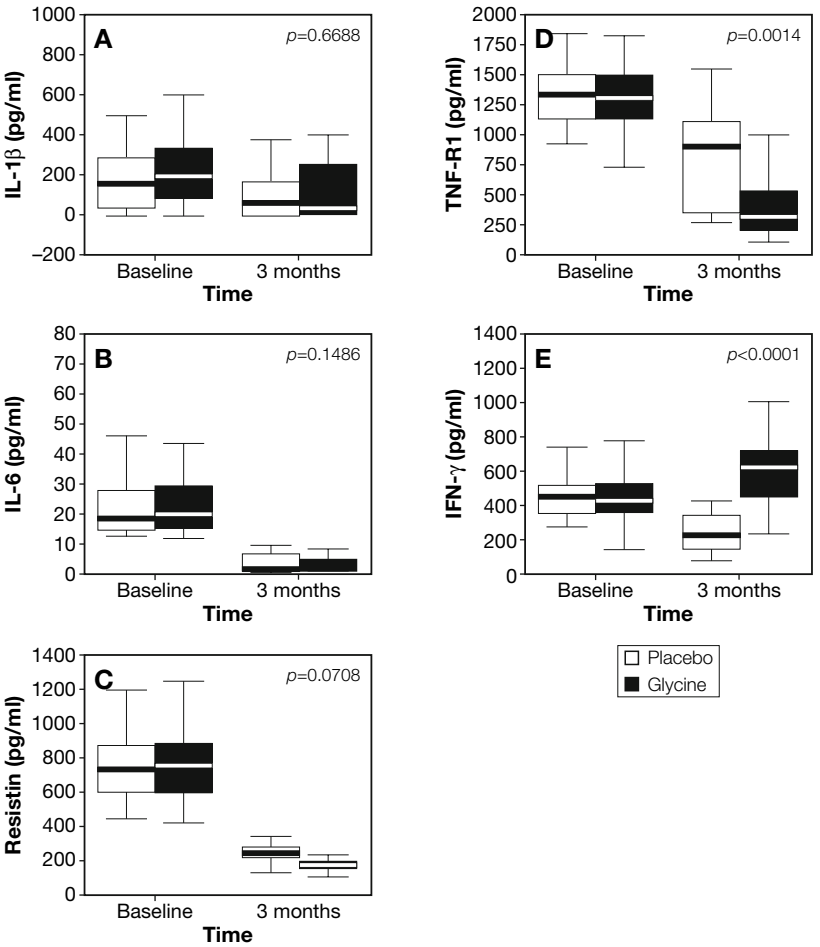

Fig. 1 - *Cytokine concentrations in patients with Type 2 diabetes after treatment with glycine or placebo. Means±SD and SE of baseline concentrations (pg/ml) and during follow-up treatment, *extreme values. (A) interleukin (IL)-1β, (B) IL-6, (C) resistin, (D) tumor necrosis factor receptor I (TNF-RI), (E) interferon (INF)-γ. p-values are for time × group interactions in a two-way analysis of variance.*

Pearson's correlations were estimated between the various cytokines. Low but statistically significant correlations were found between HOMA-IR and TNF-RI levels (r=0.237, p=0.039), and A1C and TNF-RI levels (r=0.253, p=0.002), and IFN-γ and fasting insulin levels (r=–0.234, p=0.046).

DISCUSSION

The results of this study indicate that daily ingestion of glycine, 15 g for a 3-month period, modifies several parameters in patients with Type 2 diabetes. Fasting glucose levels were reduced at 3 months, but there were no differences between groups. However, A1C declined to a greater extent in the group treated with glycine. A decrease of blood glucose reflects short-term changes in glycemic status and A1C reflects long-term glycemic status. Additionally, fasting blood glucose levels may have been subject to more variability than A1C levels, therefore there were no differences between groups. In addition, the mechanism by which A1C levels are reduced by glycine may not depend on fasting blood glucose levels alone (5). Patients and their physicians were blinded to treatment identity, and patients continued their customary treatment during the trial, therefore this does not account for group differences at the end of the 3-month treatment. No significant differences between groups were noted for fasting insulin levels and HOMA-IR. A 3-month treatment may be insufficient for glycine to affect insulin levels. Glycine and other amino acids are insulinotropic, and the effects that we observed may not be exclusive of glycine (1).

Levels of blood glucose, insulin, HOMA-IR, and all study proinflammatory cytokines were reduced during the trial. It has been previously shown that during clinical trials doctors and patients modify their customary behavior and compliance to treatment (22). Nevertheless, all analysis was adjusted for age, time since diagnosis, BMI, gender, and treatment; therefore, we do not believe our results are due to any of those factors. The results were therefore statistical differences due to the treatment with glycine.

It has been shown that levels of proinflammatory cytokines such as IL-6, TNF-α, and adipocytokines, such as resistin, increase with increasing levels of blood glucose and insulin resistance (23, 24). The correlations that we observed between TNF-RI and HOMA-IR confirm this finding. Therefore, the overall reduction in blood glucose and insulin levels and HOMA-IR may explain the overall decrease in proinflammatory cytokines in our patients.

Moreover, since these are free living individuals, a higher variability is expected than in laboratory experiments; hence the higher SD of measurements.

In this study, TNF-RI decreased to a higher degree in patients treated with glycine. It has been shown in experimental models that treatment with glycine can prevent TNF-α secretion in monocytes after an LPS stimulus (7). It has also been shown that pre-treatment with glycine prevents tissue damage in ischemia reperfusion (25). It is thought that these effects are mediated through glycine binding to two types of glycine receptor in the cell membrane, which prevents transmembrane calcium movement (20). This ultimately modulates nuclear factor (NF)-κB activation and reduces the synthesis of several proinflammatory cytokines and adhesion molecules, thus preventing endothelial damage (20, 26). We believe that this mechanism partly explains the reduction in TNF-RI levels in our patients. The correlation between A1C levels and change in TNF-RI levels confirms previous observations which showed that the NF-κB system may be activated by receptors for advanced glycation end products (RAGE) (26); and it may explain the higher decrease in TNF-RI levels in patients treated with glycine. Thus, a reduction in A1C levels may ultimately prevent organ damage and chronic complications (16). No mechanism is known for the A1C reduction in diabetes patients treated with glycine; however, at the beginning of the 20th century, German chemists added amino acids to solutions containing proteins and sugars in order to prevent the "browning" effect. This was thought to happen because the carbonyl moiety of amino acids competed with the carbonyl moieties in proteins for binding the amino moieties of sugars (20).

Several other mechanisms may explain the effects of glycine on TNF-RI levels. It has been described that glycine reduces free fatty acids, triglycerides, and oxidative stress. Moreover, glycine is a precursor of glutathione, an important antioxidant and a co-factor of glutathione peroxidasis (26). Oxidative stress is another mechanism for the activation of the NF-κB and c-Jun N-terminal kinase (JNK) systems, which are thought to activate and modulate the inflammatory response in diabetes and the metabolic syndrome (27, 28). There were no differences in triglyceride levels between groups through the trial; but we did not measure fatty acids or products of oxidative stress. Nevertheless, glycine is thought to suppress the activation of transcription factors and formation of free radicals besides the suppression of cytokine production (8).

We observed an increase in IFN-γ levels in patients treated with glycine and a decrease with placebo. There are few studies that explain these results; two studies showed that CD4+ and CD8+ T cells obtained from Type 2 diabetes patients released significantly lower amounts of IFN-γ than cells from healthy volunteers (18, 19). This observation is in agreement with our results, which show that Type 2 diabetes patients treated with placebo had lower levels of IFN-γ at the end of the study. This finding could perhaps be explained by the low proliferative response of T cells or PBMN from patients with Type 2 diabetes as occurred in diabetic rats in response to mitogens (6). However, rats treated with glycine showed an increase in the peripheral blood mononuclear cells proliferative response to mitogens (16). We believe that this increase in mitogen responsiveness can be explained by the increase in IFN-γ levels that we observed in patients treated with glycine. IFN-γ levels were negatively correlated with fasting insulin levels, suggesting that the change in IFN-γ levels will be the opposite to that of proinflammatory cytokines in Type 2 diabetes.

The increase in IFN-γ levels after glycine treatment might be of biological relevance. This cytokine is produced by activated T lymphocytes and NK cells whose principal function is to activate macrophages in both innate and adaptive immune responses, which could increase cell-mediated immunity in the patients with Type 2 diabetes.

In conclusion, our results clearly show that glycine treatment reduces levels of proinflammatory cytokines, particularly TNF-RI, and at the same time increases IFN-γ levels. This effect is likely to have a beneficial impact on innate and adaptive immune responses and could help prevent damage caused by chronic inflammation in patients with Type 2 diabetes.

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REFERENCES

1. van Loon LJ, Kruijshoop M, Menheere PP, Wagenmakers AJ, Saris WH, Keizer HA. Amino acid ingestion strongly enhances insulin secretion in patients with long-term type 2 diabetes. Diabetes Care 2003, 26: 625-30.
2. González-Ortiz M, Medina-Santillán R, Martinez-Abundis E, von Drateln CR. Effect of glycine on insulin secretion and action in healthy first-degree relatives of type 2 diabetes mellitus patients. Horm Metab Res 2001, 33: 358-60.
3. Gannon MC, Nuttall JA, Nuttall FQ. The metabolic response to ingested glycine. Am J Clin Nutr 2002, 76: 1302-7.
4. Alvarado-Vásquez N, Zamudio P, Cerón E, et al. Effect of glycine in streptozotocin-induced diabetic rats. Comp Biochem Physiol C Toxicol Pharmacol 2003, 134: 521-7.
5. Carvajal-Sandoval G, Juárez E, Ramos Martínez G, Carvajal-Juárez ME, Medina-Santillán R. Inhibition of hemoglobulin glycation with glycine induced diabetes mellitus in rats. Proc West Pharmacol Soc 1999, 42: 35-6.
6. Alvarado-Vásquez N, Lascurain R, Cerón E, et al. Oral glycine administration attenuates diabetic complications in streptozotocin-induced diabetic rats. Life Sci 2006, 79: 225-32.
7. Spittler A, Reissner CM, Oehler R. Immunomodulatory effects of glycine on LPS-treated monocytes: reduced TNF-alpha production and accelerated IL-10 expression. FASEB J 1999, 13: 563-71.
8. Zhong Z, Wheeler MD, Li X, et al. L-Glycine: a novel anti-inflammatory, immunomodulatory and cytoprotective agent. Curr Opin Clin Metab Care 2003, 6: 229-40.
9. Duncan BB, Schmidt MI, Pankow JS, et al. Low-grade systemic inflammation and the development of type 2 diabetes: the atherosclerosis risk in communities study. Diabetes 2003, 52: 1799-805.
10. Tripathy D, Mohanty P, Dhindsa S, et al. Elevation of free fatty acids induce inflammation and impairs vascular reactivity in healthy subjects. Diabetes 2003, 52: 2882-7.
11. Chu NF, Spiegelman D, Rifai N, Hotamisligil GS, Rimm EB. Glycemic status and soluble tumor necrosis factor receptor levels in relation to plasma leptin concentrations among normal weight and overweight US men. Internat J Obes Relat Metabol Disor 2000, 24: 1085-92.

12. Laaksonen DE, Niskanen L, Nyyssönen K, et al. C-reactive protein and the development of the metabolic syndrome and diabetes in middle-aged men. Diabetologia 2004, 47: 1403-10.
13. Müller S, Martin S, Koenig W, et al. Impaired glucose tolerance is associated with increased serum concentrations of interleukin 6 and co-regulated acute-phase proteins but not TNF-alpha or its receptors. Diabetologia 2002, 45: 805-12.
14. Fernández-Real JM, Broch M, Ricart W, et al. Plasma levels of the soluble fraction of tumor necrosis factor receptor 2 and insulin resistance. Diabetes 1998, 47: 1757-62.
15. Dzienis-Straczkowska S, Straczkowski M, Szelachowska M, Stepien A, Kowalska I, Kinalska I. Soluble tumor necrosis factor-alpha receptors in young obese subjects with normal and impaired glucose tolerance. Diabetes Care 2003, 26: 875-80.
16. Hu FB, Meigs JB, Li TY, Rifai N, Manson JE. Inflammation markers and risk of developing type 2 diabetes in women. Diabetes 2004, 53: 693-700.
17. Weyer C, Yudkin JS, Stehouwer CD, Schalkwijk CG, Pratley RE, Tataranni PA. Humoral markers of inflammation and endothelial dysfunction in relation to adiposity and in vivo insulin action in Pima Indians. Atherosclerosis 2002, 161: 233-42.
18. Tsiavou A, Degiannis D, Hatziagelaki E, Koniavitou K, Raptis SA. Intracellular IFN-gamma production and IL-12 serum levels in latent autoimmune diabetes of adults (LADA) and in type 2 diabetes. J Interferon Cytokine Res 2004, 24: 381-7.
19. Tsiavou A, Hatziagelaki E, Chaidaroglou A, Koniavitou K, Degiannis D, Raptis SA. Correlation between intracellular interferon-gamma (IFN-gamma) production by CD4+ and CD8+ lymphocytes and IFN-gamma gene polymorphism in patients with type 2 diabetes mellitus and latent autoimmune diabetes of adults (LADA). Cytokine 2005, 31: 135-41.
20. El Hafidi M, Pérez I, Baños G. Is Glycine effective against elevated blood pressure? Curr Opin Clin Nutr Metab Care 2006, 9: 26-31.
21. Matthews DR, Hosker JP, Rudenski AS, Naylor BA, Treacher DF, Turner RC. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia 1985, 28: 412-9.
22. Andrade SE, Walker AM, Gottlieb LK, et al. Discontinuation of antihyperlipidemic drugs-do rates reported in clinical trials reflect rates in primary care settings? N Engl J Med 1995, 332: 1125-31.
23. Esposito K, Nappo F, Marfella R, et al. Inflammatory cytokine concentrations are acutely increased by hyperglycemia in humans: role of oxidative stress. Circulation 2002, 106: 2067-76.
24. Kusminski CM, McTernan PG, Kumar S. Role of resistin in obesity, insulin resistance and Type II diabetes. Clin Sci (Lond) 2005, 109: 243-56.
25. Slomowitz LA, Deng A, Hammes JS, Gabbai F, Thomson SC. Glomerulotubular balance, dietary protein, and the renal response to Glycine in diabetic rats. Am J Physiol Regul Integr Comp Physiol 2002, 282: R1096-103.
26. Zelvyte I, Dominaitiene R, Crisby M, Janciauskiene S. Modulation of inflammatory mediators and PPARgamma and NF-kappaB expression by pravastatin in response to lipoproteins in human monocytes in vitro. Pharmacol Res 2002, 45: 147-54.
27. Matilla B, Mauriz JL, Culebras JM, Gonzalez-Gallego, Gonzalez P. La glicina: un antioxidante protector celular. Nutr Hosp 2002, 1: 2-9.
28. Bierhaus A, Schiekofer S, Schwaninger M, et al. Diabetes-associated sustained activation of transcription factor nuclear factor-kappa B. Diabetes 2001, 50: 2792-808.