Differential Effect of the Antidiabetic Thiazolidinediones Troglitazone and Pioglitazone on Human Platelet Aggregation Mechanism

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Troglitazone and pioglitazone, antidiabetic thiazolidinediones, are known to improve insulin resistance. However, the effect of these drugs on platelet aggregation remains unclear. The chemical structure of troglitazone contains vitamin E. Accordingly, we studied the effect of troglitazone, pioglitazone, and vitamin E on thrombin-induced platelet aggregation, metabolism of phosphoinositide, protein phosphorylation, protein kinase C (PKC)-α and -β, and phosphatidylinositol (PI) 3-kinase activation in vitro in human platelets. Maximum platelet aggregation by ADP, collagen, and thrombin decreased in the presence of 0.1–1 μmol/l troglitazone and 500 nmol/l vitamin E for 60 min compared with controls. However, pioglitazone did not inhibit ADP-, collagen-, or thrombin-induced platelet aggregation. Pretreatment with troglitazone and vitamin E, but not with pioglitazone, resulted in decreases in thrombin-induced phosphatidic acid production, hydrolysis of phosphatidylinositol 4,5-bisphosphate by phospholipase C, and 47-kDa protein phosphorylation. Thrombin-induced PKC-α and -β activation in membrane fraction was suppressed by pretreatment with troglitazone and vitamin E, but not with pioglitazone. Separately, troglitazone and pioglitazone stimulated PI 3-kinase activity, but thrombin-induced PI 3-kinase activation was suppressed by pretreatment with troglitazone and pioglitazone for 60 min. These results suggest that troglitazone and vitamin E, but not pioglitazone, have a potent inhibitory effect on platelet aggregation via suppression of the thrombin-induced activation of phosphoinositide signaling in human platelets. Finally, the chemical structure of vitamin E may contribute to the inhibitory effect of troglitazone on platelet aggregation in human platelets. *Diabetes* 47:1494–1500, 1998

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DG, diacylglycerol; HPTLC, high-performance thin-layer chromatography; PA, phosphatidic acid; PI, phosphatidylinositol; PIP, phosphatidylinositol-4-monophosphate; PIP_2, phosphatidylinositol 4,5,bisphosphate; PKC, protein kinase C; PLC, phospholipase C; TPA, 12-*o*-tetradecanoylphorbol-13-acetate.

Platelet functions such as aggregation, serotonin and ADP secretion, and thromboxane-A_2 production are mediated by some signal transduction mechanism(s). Thrombin causes hydrolysis of phosphatidylinositol 4,5,bisphosphate (PIP_2) by phospholipase C (PLC), with concomitant formation of diacylglycerol (DG) and inositol 1,4,5-triphosphate, which induces a rapid release of Ca^{2+} from a dense tubular system (1–5) and stimulates phospholipase-A_2 to release arachidonic acid. DG is known to activate protein kinase C (PKC) (6,7), and is effectively phosphorylated to phosphatidic acid (PA), which can then be redirected to phosphatidylinositol (PI) through cytidine diphosphate–DG. Arachidonic acid is oxygenated to produce thromboxane-A_2 (8). Platelet aggregation is increased in diabetic patients compared with in healthy subjects (9,10). Moreover, enhanced arachidonic acid release and thromboxane production have been reported in platelets from NIDDM patients (11,12). Recently, a few reports (13–15) focused on thrombin-induced changes in phosphoinositide metabolism in diabetic patients. Decreased platelet phosphoinositide turnover and enhanced platelet activation have been reported in IDDM patients (13). On the other hand, the thiazolidinedione troglitazone acts by enhancing the action of insulin without stimulating insulin secretion from the pancreas (16–18). However, the mechanism that induces this enhancement in insulin action is unclear. Moreover, there is no information regarding the effect of thiazolidinediones on the platelet aggregation mechanism. Troglitazone contains the chemical structure of vitamin E, but not pioglitazone.

In this study, we evaluated the differential effects of thiazolidinediones such as troglitazone and pioglitazone and vitamin E on thrombin-induced platelet aggregation, phosphoinositide metabolism, protein phosphorylation, and PI 3-kinase activation in human platelets.

RESEARCH DESIGN AND METHODS

Materials. High-performance thin-layer chromatography (HPTLC) plates were purchased from Merck (Rahway, NJ). [^{32}P]orthophosphate was obtained from Du Pont-NEN (Boston, MA). Thrombin (bovine), ADP, collagen, 12-*o*-tetradecanoylphorbol-13-acetate (TPA), phosphatidylinositol-4-monophosphate (PIP), and PIP_2 as standards were obtained from Sigma (St. Louis, MO). α-Tocopherol acetate was donated by Eizai Pharmaceutical (Tokyo, Japan), troglitazone was donated by Sankyo Pharmaceutical (Tokyo, Japan), and pioglitazone was donated by Takeda Pharmaceutical (Osaka, Japan). All other chemicals used in this study were of reagent grade.

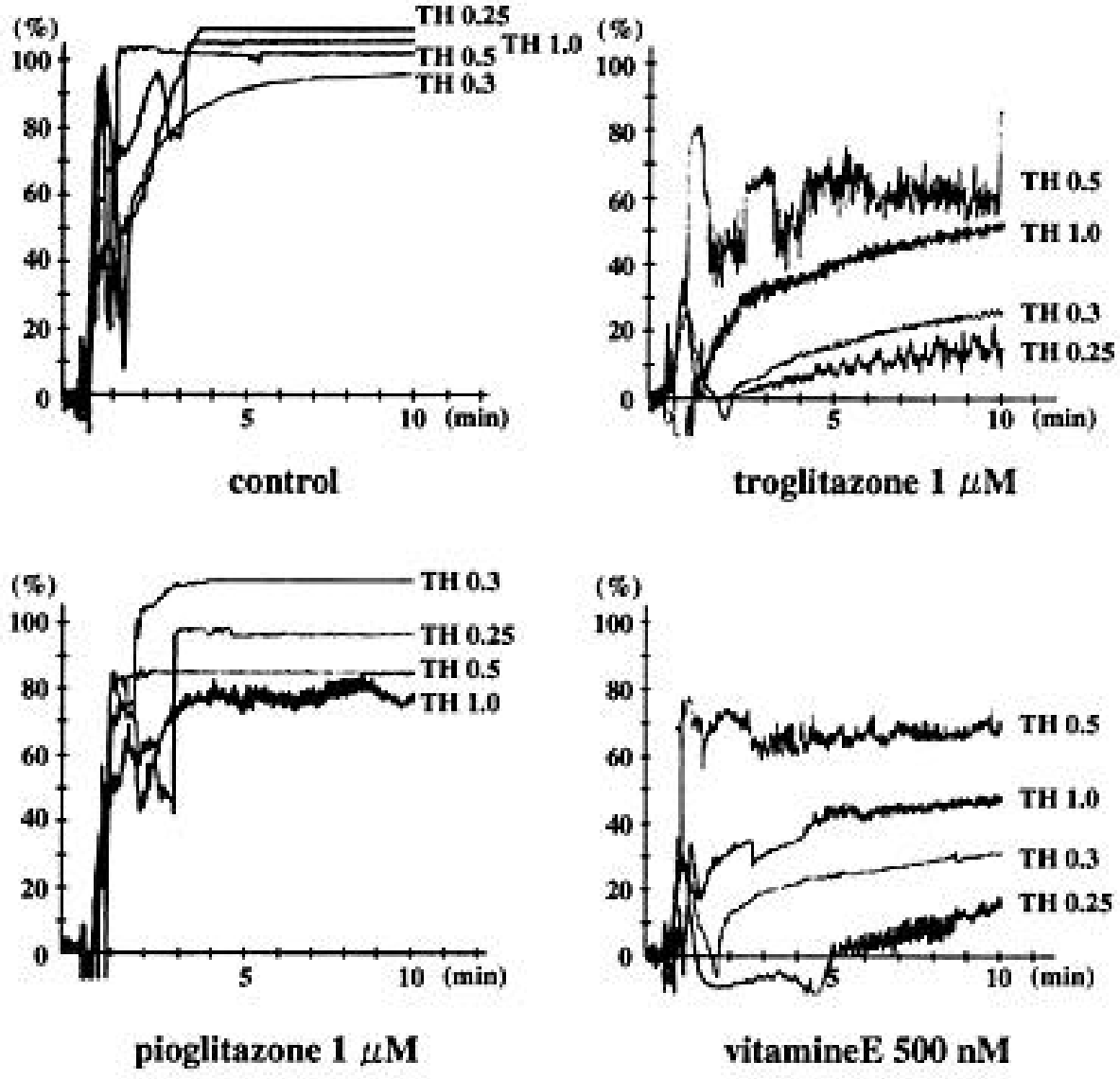

FIG. 1. Effect of troglitazone, pioglitazone, and vitamin E on thrombin (TH)-induced platelet aggregation. Platelet aggregation was achieved by Born's turbidimetric method with an aggregometer. Preincubation with 1 μmol/l troglitazone and 500 nmol/l vitamin E, but not 1 μmol/l pioglitazone, for 60 min decreased thrombin-induced platelet aggregation. Each panel shows a thrombin-stimulated (0.25–1 U/ml) aggregation profile, as shown by an aggregometer without (control) or with thiazolidinedione or vitamin E treatment. The same results were obtained from four other experiments.

Preparation of [^{32}P]-labeled human platelets. Fresh blood was obtained from control subjects with their consent, and then centrifuged at 164*g* for 10 min, as previously described (15). The resulting supernatant (platelet-rich plasma) was centrifuged at 800*g* for 10 min, washed, resuspended in buffer I (15.4 mmol/l Tris, 140 mmol/l NaCl, 5.6 mmol/l glucose [pH 7.4]), and incubated with [^{32}P]orthophosphate (200 mCi) for 90 min at 37°C. The labeled platelets were washed and resuspended in buffer I containing 1 mmol/l $CaCl_2$ at a final concentration of 10^9/ml.

Platelet aggregation. Platelet aggregation was achieved using Born's turbidimetric method (19) with an aggregometer (NBS Hema Tracer 801; Niko Bioscience, Tokyo, Japan). Briefly, 100 μl of platelet-rich plasma was stirred magnetically at 1,100 rev/min at 37°C. After a 1-min incubation, 0.25, 0.3, 0.5, or 1 U/ml bovine thrombin was added, and the change in light transmission was recorded for 7 min. The maximum aggregation was determined as the peak light transmission after the addition of thrombin.

Lipid analysis. After the platelet suspension (490 μl) was incubated with 1 U/ml thrombin, 2 ml of chloroform/methanol/HCl (20:40:1, vol/vol/vol) (20) were added; lipid was then extracted by a modification of the Bligh-Dyer method (21). The resultant mixture was supplemented with 0.5 ml chloroform, mixed, and centrifuged at 2,500 revol/min for 5 min. The lower phase was collected and 0.5 ml of a mixture of 0.2 mol/l KCl and 5 mmol/l EDTA was added. The phases were separated by centrifugation, and the lower phase was collected and dried under nitrogen. The phosphoinositides were separated on HPTLC plates and impregnated with 1% potassium oxalate in a solvent system of chloroform/acetone/methanol/acetic acid/water (40:15:13:12:8, vol/vol/vol/vol/vol) (22). The areas corresponding to individual lipids, identified by co-migration with authentic standards, were scraped into vials, and radioactivity was determined by a scintillation counter with toluene/Triton X-100/water/2,2′-*p*-phenylene-bis(5-phenyloxazole)/2,5 diphenyloxazole (800 ml:200 ml:50 ml:0.24 g:3.3 g). The rate of each phospholipid radioactivity was calculated.

Phosphorylation of 47-kDa protein. ^{32}P-labeled platelets were stimulated with 1 U/ml thrombin for 1 min or 10^{-6} mol/l TPA for 1 or 10 min. The reaction was terminated by adding sample buffer (150 mmol/l Tris/HCl [pH 6.8], 7.5% SDS, 12% (vol/vol) glycerol, 4.5% 2-mercaptoethanol) and then placing the samples in boiling water for 3 min. Samples were kept overnight at 4°C and then subjected to SDS-PAGE using 12.5% acrylamide separating gels and 4.5% stacking gels, both containing 0.1% SDS. The gels were dried and exposed to Kodak X-Omatic film (Eastman-Kodak, Rochester, NY) for 72 h at –50°C to prepare autoradiographs. The intensity of the phosphorylated protein was scanned with a laser densitometer (Pharmacia LKB Biotechnology, Tokyo, Japan) to determine the relative value.

PKC experiments. Reactions were terminated by the addition of 20 mmol/l Tris-HCl buffer (pH 7.5) containing 0.25 mol/l sucrose, 1.2 mmol/l EGTA, 0.1 mmol/l phenylmethylsulfonyl fluoride, 20 μg/ml leupeptin, and 20 mmol/l 2-mercaptoethanol (buffer I); they were then washed twice and homogenized in buffer I. The homogenates were centrifuged for 60 min at 105,000*g* to obtain the cytosol and membrane fractions. The latter was homogenized in buffer I containing 5 mmol/l EGTA,

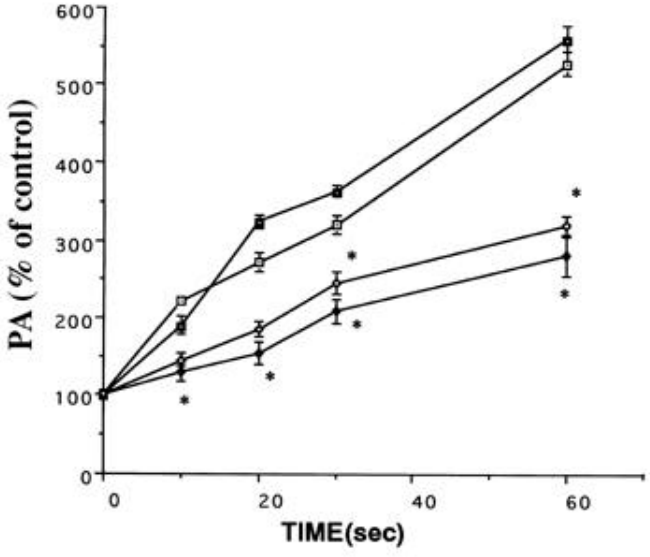

FIG. 2. Effect of troglitazone, pioglitazone, and vitamin E on thrombin-induced PA formation. [^{32}P]-labeled platelets were stimulated with 1 U/ml thrombin after preincubation without (control; □) or with 1 μmol/l troglitazone (◆), 1 μmol/l pioglitazone (■), or 500 nmol/l vitamin E (◇) for 60 min. The reaction was terminated with chloroform/methanol/HCl, and the resultant lipid was extracted as indicated in METHODS. Radioactivity of PA was counted by scintillation counter as shown in METHODS. Data are plotted as means ± SE of five separate experiments. *P < 0.05 vs. control.

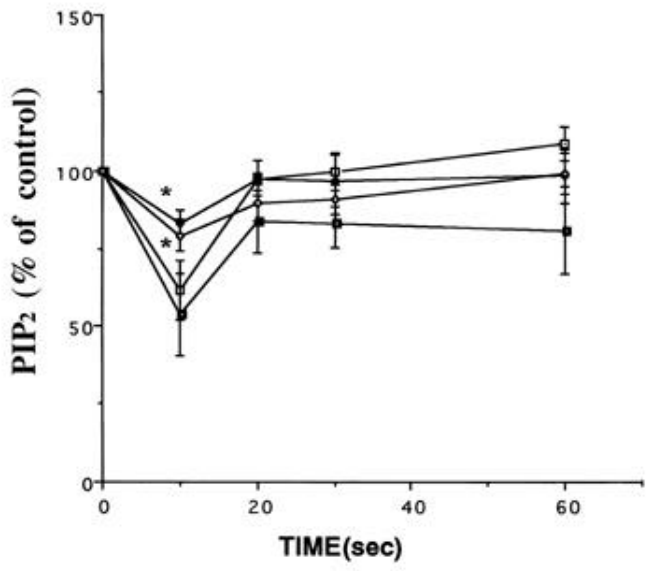

FIG. 3. Effect of troglitazone, pioglitazone, and vitamin E on thrombin-induced incorporation of [^{32}P] radioactivity into PIP_2. [^{32}P]-labeled platelets were stimulated with 1 U/ml thrombin after preincubation without (control; □) or with 1 μmol/l troglitazone (◆), 1 μmol/l pioglitazone (■), or 500 nmol/l vitamin E (◇) for 60 min. Radioactivity of PIP_2 was analyzed as shown in METHODS. Data are plotted as means ± SE of five separate experiments. *P < 0.05 vs. control.

2 mmol/l EDTA, and 1% Triton X-100. Activation of PKC in platelets was assayed by changes in the subcellular distribution of immunoreactive PKC, using the methods previously described (23,24). Equal amounts of cytosol or membrane-associated fraction were prepared, subjected to SDS-PAGE, transferred to nitrocellulose, and incubated first with polyclonal antiserum raised to synthetic peptide to PKC-α and -β (Gibco, Grand Island, NY) and second with goat anti-rabbit globulin complexed to alkaline phosphatase (Sigma) or an enhanced chemiluminescence system (Amersham, Tokyo, Japan). This immunoblotting method detected a single major immunoreactive band that co-migrated on SDS-PAGE and blotted identically with 80-kDa (PKC-β) synthetic peptides.

PI 3-kinase activity. After preincubation with or without 1 μmol/l troglitazone, platelets were treated with 1 U/ml thrombin for the indicated periods and lysed in a buffer (20 mmol/l Tris/HCl [pH 7.4], 0.5% Nonident P-40, 0.5 mmol/l EDTA, 5% glycerol, 1 mmol/l orthovanadate, and 20 μmol/l *p*-amidinophenylmethanesulphonyl fluoride hydrochloride) and sonicated. The homogenate was subjected to a PI kinase assay in a 50-μl reaction mixture containing 20 mmol/l Tris/HCl [pH 7.4], 100 mmol/l NaCl, 10 mmol/l $MgCl_2$, 0.5 mmol/l EGTA, 100 μmol/l phosphatidylserine, and 10 μmol/l [γ-^{32}P]ATP (0.1 μCi/μl). After 10 min at 30°C, the reaction was stopped by adding 200 μl of 1 mol/l HCl and 80 μl of chloroform/methanol (1:1, vol/vol). A 30-μl portion of the lower layer was spotted onto a Silica Gel 60 plate (Merck) and developed in chloroform/methanol/28% NH_4Cl/water (70:100:15:25, vol/vol/vol/vol) (25). The radioactive PI phosphate spot was detected by autoradiography, scraped from the plate, and quantified by liquid scintillation counting.

Statistical analysis. Statistical analysis was performed by Student's t test. Statistical significance was defined as $P < 0.05$.

RESULTS

Effect of thiazolidinediones and vitamin E on platelet aggregation. Various concentrations (0.25–1 U/ml) of thrombin-stimulated aggregation were suppressed by pretreatment with 1 μmol/l troglitazone, 1 μmol/l pioglitazone, or 500 nmol/l vitamin E. (Note that this concentration was less than the maximal plasma concentration when used as a therapeutic dose. The maximum concentrations of troglitazone, pioglitazone, and α-tocopherol acetate were 1.36, 3.8, and 8.5 μmol/l, respectively.) In particular, a low concentration (0.25 or 0.3 U/ml) of thrombin-stimulated maximum aggregation was significantly decreased compared with in the controls (0.1–0.01% dimethyl sulfoxide), as indicated in Fig. 1. ADP- or collagen-stimulated platelet aggregation was also suppressed by pretreatment with troglitazone (data not shown).

Effect of thiazolidinediones and vitamin E on thrombin-induced phosphoinositide metabolism. When platelets were labeled with [^{32}P]orthophosphate, thrombin-induced production of [^{32}P]PA after pretreatment with 1 μmol/l troglitazone or 500 nmol/l vitamin E for 60 min significantly decreased for 10, 20, 30, and 60 s compared with those without thiazolidinedione pretreatment (control) (Fig. 2). Pretreatment with 1 μmol/l pioglitazone resulted in no significant difference in thrombin-induced PA production from that of controls. Thrombin-induced changes of [^{32}P]PI and PIP after pretreatment with thiazolidinediones and vitamin E were not significantly different from controls (data not shown). On the other hand, thrombin-induced incorporation of [^{32}P] radioactivity into PIP_2 for 10 s after pretreatment with 1 μmol/l troglitazone or 500 nmol/l vitamin E was significantly higher than in controls. However, pretreatment with 1 μmol/l pioglitazone demonstrated no significant difference compared with controls (Fig. 3). These results suggest that troglitazone and vitamin E, but not pioglitazone, suppress thrombin-induced PLC activation.

Effect of thiazolidinediones and vitamin E on thrombin- or TPA-induced 47-kDa protein phosphorylation. Thrombin- and TPA-stimulated 47-kDa protein phosphorylation in [^{32}P]-labeled platelets significantly increased for 10 min (Fig. 4, *lanes 2* and *3*). Pretreatment with 100 nmol/l (*lanes 4* and *5*) or 1 μmol/l troglitazone (*lanes 6* and *7*)

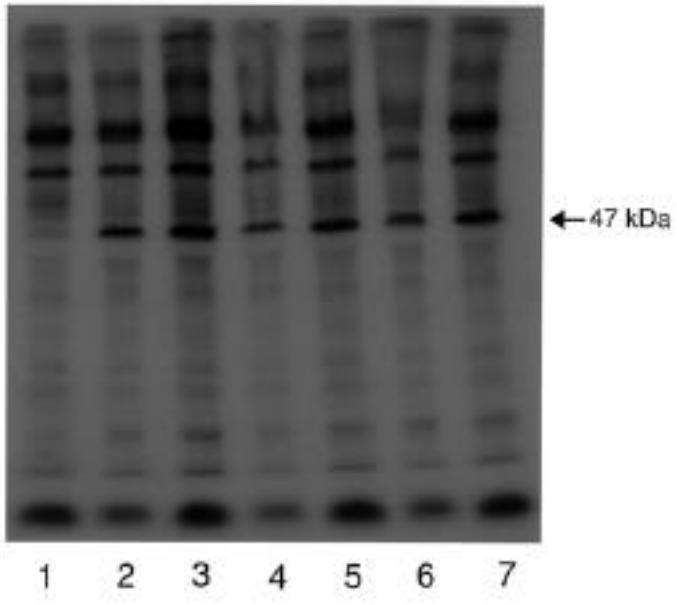

FIG. 4. Effect of troglitazone on thrombin- or TPA-induced 47-kDa protein phosphorylation. [^{32}P]-labeled platelets were stimulated without (control; *lane 1*) or with 1 U/ml thrombin (*lane 2*) or 1 µmol/l TPA (*lane 3*) for 10 min after pretreatment with 100 nmol/l (*lanes 4* and *5*) or 1 µmol/l (*lanes 6* and *7*) troglitazone. Thrombin- (*lanes 4* and *6*) and TPA-induced (*lanes 5* and *7*) 47-kDa protein phosphorylation for 10 min after pretreatment with troglitazone is shown in *lanes 4–7*. A representative experiment is shown here. Similar results were obtained from four other experiments.

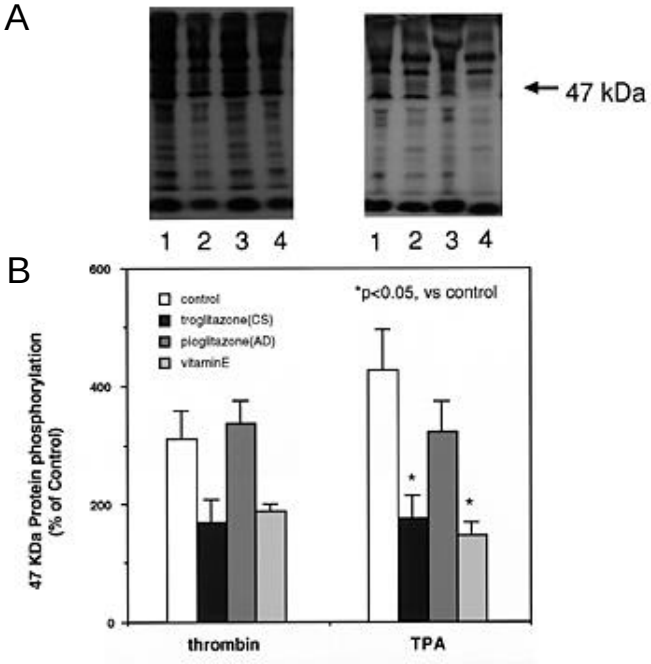

FIG. 5. Effect of thiazolidinedione and vitamin E on thrombin- and TPA-induced 47-kDa protein phosphorylation in human platelet. [^{32}P]-labeled platelets were pretreated without (control; *lane 1*) or with 1 µmol/l troglitazone (*lane 2*), 1 µmol/l pioglitazone (*lane 3*), or 500 nmol/l vitamin E (*lane 4*) for 60 min. Then 1 U/ml thrombin-stimulated (*left panel*) or 1 µmol/l TPA-stimulated (*right panel*) 47-kDa protein phosphorylations for 10 min were measured as shown in *A*, which is a representative experiment. Protein phosphorylation was analyzed as shown in METHODS. Each value indicated 47-kDa protein phosphorylation by densitometric analysis as a percentage of the control (without stimulation: 100%). Data are plotted as means ± SE of five separate experiments in *B*. *P < 0.05 vs. control.

resulted in decreases in thrombin- and TPA-stimulated 47-kDa protein phosphorylation (Fig. 4, *lanes 4–7*). As shown in Fig. 5*A*, thrombin- and TPA-induced 47-kDa protein phosphorylations for 10 min after pretreatment with 1 µmol/l troglitazone (*lane 2*) or 500 nmol/l vitamin E (*lane 4*) for 60 min were significantly suppressed compared with in controls, but not after pretreatment with 1 µmol/l pioglitazone (*lane 3*). Laser densitometric analysis showed that troglitazone and vitamin E pretreatment significantly (P < 0.05) decreased TPA-induced 47-kDa protein phosphorylation (Fig. 5*B*).

Effect of thiazolidinediones and vitamin E on thrombin-induced PKC-α and -β activations. To clarify the inhibitory effect of thiazolidinediones and vitamin E on thrombin-induced conventional PKC activation, PKC-α and -β of platelets in membrane fraction were examined after pretreatment of those agents. As shown in Fig. 6, thrombin-induced PKC-α and -β immunoreactivities in membrane fraction for 10 min were decreased in the presence of troglitazone and vitamin E, but not pioglitazone, for 60 min compared with controls. Cytosolic PKC-α and -β immunoreactivities were slightly increased in the presence of troglitazone and vitamin E, but not pioglitazone, compared with controls. (Note that the cytosolic protein content was significantly higher than the membrane content [data not shown].)

Dosage-responsive effect of thrombin on PI 3-kinase activity. Various concentrations (0.01–1 U/ml) of thrombin stimulated PI 3-kinase activity for 1 min under thin-layer chromatography. As shown in Fig. 7, dosage-responsive increases of PI 3-kinase activity could be observed.

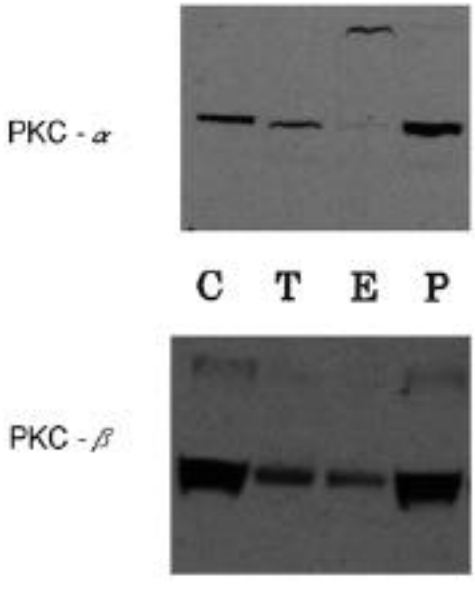

FIG. 6. Effect of thiazolidinediones and vitamin E on thrombin-induced PKC-α and -β activations. Platelet suspension was treated without (control; C) or with 1 µmol/l troglitazone (T), 500 nmol/l vitamin E (E), or 1 µmol/l pioglitazone (P) for 60 min, and then stimulated with 1 U/ml thrombin for 10 min. Each sample was sonicated and centrifuged to obtain cytosol and membrane fraction. Equal amounts of membrane-associated protein (30 µg) were analyzed by immunoblotting. PKC-α and -β immunoreactivities were measured as shown in METHODS. A representative experiment is shown here. Similar results were obtained from three other experiments.

FIG. 7. Thrombin-induced PI 3-kinase activation. Platelets were stimulated with 0.01–1 U/ml thrombin for 1 min, and then sonicated in lysis buffer. Resultant homogenate was subjected to PI kinase assay as indicated in METHODS. A representative experiment is shown here. Similar results were obtained from four other experiments. PI3P, phosphatidylinositol 3 phosphate.

Effect of thiazolidinediones and vitamin E on thrombin-induced PI 3-kinase activation. Separately, troglitazone and pioglitazone stimulated PI 3-kinase activity. Thrombin-induced increase of PI 3-kinase activity after pretreatment with 100 nmol/l or 1 µmol/l troglitazone was significantly suppressed compared with in controls (i.e., those without troglitazone pretreatment), as shown in Fig. 8. Pretreatment with 1 µmol/l pioglitazone, but not with vitamin E, also suppressed an increase in thrombin-induced PI 3-kinase activity.

DISCUSSION

We previously demonstrated how sulfonylureas suppress the platelet aggregation mechanism (26); sulfonylureas suppress platelet PLC, PA, and DG production, resulting in 47-kDa protein phosphorylation by PKC. Recently it has been reported that wortmannin inhibits irreversible platelet aggregation, which is the conversion of integrin $\alpha IIb\beta_3$ into a fibrinogen-binding form required for platelet aggregation via inhibition of thrombin-induced PI 3-kinase activation (27,28). Therefore, the platelet aggregation mechanism is influenced by PKC and PI 3-kinase activations. In this study, we investigated the effect of two new antidiabetic agents, troglitazone and pioglitazone, on the platelet aggregation mechanism concerned with phosphoinositide metabolism and PKC and PI 3-kinase activation in human platelet in vitro. Moreover, troglitazone contains the same chemical structure as that found in vitamin E. In this study, we focused on the differences in chemical structure of representative thiazolidinediones, troglitazone and pioglitazone. Accordingly, we examined the effect of vitamin E on the platelet aggregation mechanism.

As is shown in Figs. 1–3 , troglitazone, like vitamin E, suppressed thrombin-induced platelet aggregation and PLC activation, which is shown as PA production, whereas pioglitazone did not. In fact, thrombin- and TPA-induced 47-kDa protein phosphorylation was suppressed by troglitazone and vitamin E (Figs. 4 and 5). Moreover, thrombin-induced PKC

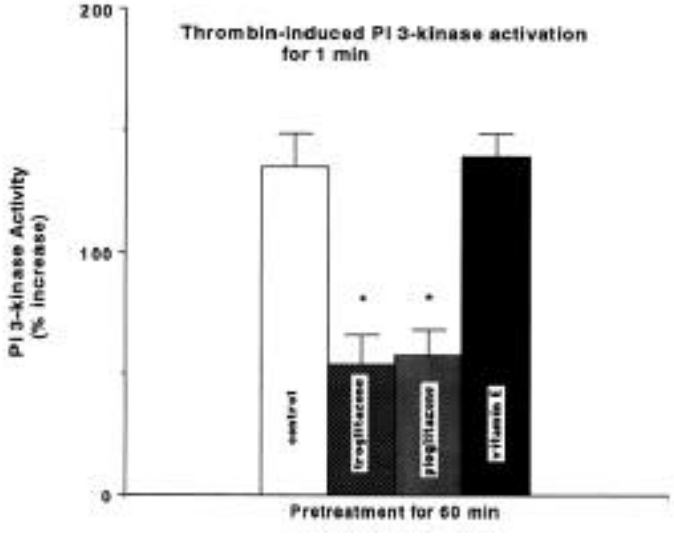

FIG. 8. Effect of thiazolidinediones and vitamin E on thrombin-induced PI 3-kinase activation. Platelets were stimulated without (control) or with 1 U/ml thrombin for 1 min after pretreatment without (control) or with 1 µmol/l troglitazone, 1 µmol/l pioglitazone, or 500 nmol/l vitamin E, and then sonicated in lysis buffer. PI 3-kinase activities were measured as shown in METHODS. PI 3-phosphate spots were scanned by laser densitometer. Data are plotted as means ± SE of five different experiments. *P < 0.05 vs. control.

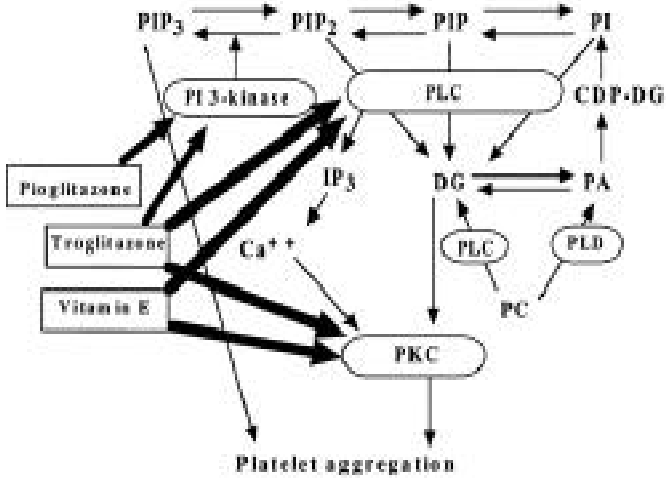

FIG. 9. Effect of thiazolidinediones and vitamin E on polyphosphatidylinositol and PKC signaling pathways in human platelets. The PI cycle has PI and DG as common metabolites; polyphosphoinositide and PI 3-kinase are believed to be controlled by thrombin stimulation. The → indicates inhibitory actions of thiazolidinediones and vitamin E on PLC, PKC, and PI 3-kinase. CDP-DG, cytidine diphosphate diacylglycerol; IP_3, inositol 1,4,5-triphosphate; PC, phosphatidylcholine; PIP_3, phosphatidylinositol 3,4,5-triphosphate; PLD, phospholipase D.

activation was decreased in the presence of troglitazone and vitamin E, but not pioglitazone, compared with controls (Fig. 6). On the other hand, thrombin-stimulated PI 3-kinase activation was inhibited by pretreatment with troglitazone and pioglitazone (Fig. 8). Both troglitazone and pioglitazone by themselves, but not vitamin E, may increase PI 3-kinase activity during pretreatment with thiazolidinediones, and subsequent PI 3-kinase activation would then be desensitized and suppressed by thrombin stimulation (Fig. 8). It should be emphasized that thrombin-induced PI 3-kinase activation can be inhibited by either troglitazone or pioglitazone treatment, but only troglitazone treatment inhibits platelet aggregation. It may be possible that thrombin-stimulated hydrolysis of phosphoinositide principally mediates a primary phase of aggregation, whereas PI 3-kinase activation is necessary for prolonged GPIIb–IIIa activation and irreversible platelet aggregation (27). Pioglitazone-mediated inhibition of PI 3-kinase activation may be weak in its suppression of platelet aggregation, whereas troglitazone, containing a chemical structure similar to that of vitamin E, strongly inhibits platelet aggregation.

Recently pioglitazone has been shown to promote insulin-induced activation of PI 3-kinase in 3T3-L1 adipocytes by inhibiting a negative control mechanism through cyclic AMP accumulation (29). Moreover, pioglitazone and other thiazolidinediones, such as AD-5075 and CP-86325, potentiate insulin-stimulated PI 3-kinase activation in insulin receptor–overexpressing CHO cells (30). The above results with regard to insulin signal transduction differ considerably from thrombin-induced signal transduction in human platelets.

One recent interesting finding was that α-tocopherol (vitamin E) inhibits aggregation of human platelets by a PKC-dependent mechanism (31). Furthermore, it has been reported that vitamin E can inhibit cell proliferation via suppression of PKC activity (32), and that retinal blood flow can be increased by the suppression of the DG-PKC signal in a vitamin E–treated diabetic rat (33). These reports suggest that vitamin E inhibits hyperglycemia-mediated DG-PKC activation. Troglitazone includes the same chemical structure as that found in vitamin E. Based on the above findings, it is reasonable to assume that troglitazone, but not pioglitazone, inhibits thrombin-induced platelet aggregation, 47-kDa protein phosphorylation, and PKC-α and -β activation in a manner similar to vitamin E in human platelet. Finally, efficacious sites of these representative thiazolidinediones—troglitazone and pioglitazone—and vitamin E on thrombin-induced signal transduction pathways are shown in Fig. 9. However, further studies regarding whether in vivo treatment with troglitazone affects the human platelet aggregation mechanism and may prevent the development of diabetic complications and atherosclerosis are required.

In conclusion, a new antidiabetic agent, troglitazone, unlike pioglitazone, has been found to decrease thrombin-induced platelet aggregation, PA production, 47-kDa protein phosphorylation, and activation of PKC-α and -β and PI 3-kinase. Based on the results of this study, troglitazone may prove to be efficacious in preventing diabetic complications and atherosclerosis in future diabetic treatment.

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