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Species-Specific Hypoglycemic Activity of Triphenylphosphoranylideneacetophenones (39184)

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(Introduced by J. H. Leathem)

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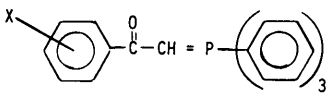
In evaluating compounds for hypoglycemic activity, it was discovered that 2-triphenylphosphoranylideneacetophenones are hypoglycemic in various rat models, but inactive in guinea pigs, rabbits, and dogs. This paper presents the biologic activity of two of these compounds.

Materials and methods. The compounds investigated in these experiments were 2 - triphenylphosphoranylideneacetophenone (SK&F 45359, X = H, 2-TPA) and 2-triphenylphosphoranylidene-*m*-trifluoromethyl-acetophenone (SK&F 62775, X = *m*-CF_3, 2-TPTA).



Normal male guinea pigs and rats and alloxan diabetic rats were handled as previously described (1), except that the criterion for diabetes was an 18-hr fasting blood glucose concentration greater than 200 mg/100 ml.

Male New Zealand white rabbits, weighing approximately 2 kg, were maintained on Tekland rabbit diet until 4 hr before use, when all food was removed. Water was provided *ad libitum*. Male and female mongrel dogs, maintained on alternate days of Wayne Dog Blocs and Kennel Ration, were fasted overnight, and weighed 11.6–15.2 kg. Water was provided *ad libitum*.

Blood glucose was determined by the glucose oxidase–Perid method (Boehringer-Mannheim) or the microferricyanide procedure for the Technicon AutoAnalyzer on tail vein samples from rats, stump blood samples from decapitated guinea pigs, ear vein samples from rabbits, and jugular vein samples from dogs.

Insulin was determined in serum samples frozen at −11° until assayed by the radioimmune assay (Schwarz/Mann insulin RIA kit).

For rats and guinea pigs, the compounds were suspended in 0.5% (w/v) aqueous gum tragacanth and administered orally by intubation. For rabbits, the compounds were administered intravenously, dissolved in 80% (v/v) aqueous ethanol. Control animals received equal volumes of the appropriate vehicle.

Dogs were orally administered gelatin capsules containing the compound. Each dog served as its own control with post-treatment values being compared to pre-treatment values.

Statistical calculations were done by Student's *t* test.

Results. Figure 1 depicts the hypoglycemic effect of 2-TPA in fed, 48- and 18-hr-fasted alloxan diabetic rats following a 150 mg/kg oral dose. Within the time frame studied, the maximum hypoglycemic response occurred at 4 hr. The alloxan diabetic rat appeared to be more susceptible, and the fed rat the least susceptible, to the hypoglycemic action of the compound.

Figure 2 depicts the results obtained with the trifluoromethyl analog (2-TPTA) in the fed, 48- and 18-hr-fasted alloxan diabetic rats. Trifluoromethyl substitution appears to endow the compound with greater hypoglycemic activity, since the results shown were obtained with an oral dose of 100 mg/kg. Again, the diabetic rat appeared to be more susceptible to the action of the compound, while the fasted and fed rats responded to a similar degree.

Figure 3 shows the dose response obtained with 2-TPA and 2-TPTA in the fed rat after the oral administration of 37.5, 75, and 150 mg/kg. Both compounds elicited a greater hypoglycemic response with increasing doses. In this experiment, the trifluoro-

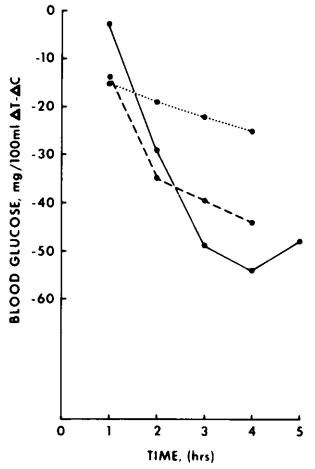


FIG. 1. Effect of 2-TPA on blood glucose of fed (●·····●), 48-hr-fasted (●- - -●), and alloxan diabetic (●——●) rats. The results are plotted as the difference between the mean blood glucose values of the controls and treated rats. Seven rats per group for fed and fasted, and 11 per group for alloxan diabetic.

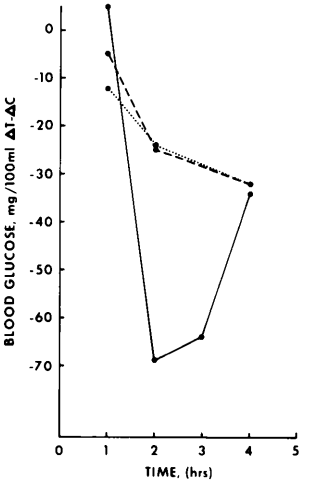


FIG. 2. Effect of 2-TPTA on blood glucose of fed (●·····●), 48-hr-fasted (●- - -●), and alloxan diabetic (●——●) rats. The results are plotted as in Fig. 1. Seven rats per group for fed and fasted, and 11 per group for alloxan diabetic.

methyl analog did not appear to possess a greater degree of activity.

To determine if the hypoglycemia was accompanied by alteration in serum insulin levels, 48-hr-fasted rats were administered 2-TPA, 150 mg/kg orally, and killed 4 hr later after obtaining a tail vein blood sample. Stump blood was collected for the determination of serum insulin concentrations. The results are shown in Table I. Four hours post-treatment, the blood glucose in the treated rats was significantly lower than that of the controls, whereas the serum insulin concentration was slightly, but statistically significantly, elevated.

To ascertain whether the hypoglycemia could possibly be due, in part, to an increased glycogenesis, 48-hr-fasted rats were administered 2-TPA, 75 and 150 mg/kg orally, and sacrificed 4 hr later, and hepatic glycogen determined. The results are shown in Table I. Both doses caused a statistically significant decrease in blood glucose during the 4-hr period; however, only the 150 mg/kg dose caused a slight, but statistically significant, increase in hepatic glycogen.

In an attempt to demonstrate hypogly-

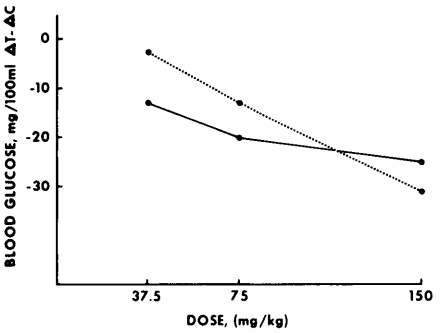


FIG. 3. Dose response of 2-TPA (●——●) and 2-TPTA (●-----●) in fed rats. The results are plotted as in Fig. 1. Seven rats per group.

TABLE I. EFFECT OF 2-TRIPHENYLPHOSPHORANYLIDENEACETOPHENONE (2-TPA) ON BLOOD GLUCOSE, SERUM INSULIN, AND HEPATIC GLYCOGEN OF 48-HOUR FASTED RATS

Treatment[a]	Blood glucose[b] (mg/100 ml)	Hepatic glycogen[b] (mg/g)	Serum insulin[b] (μU/ml)
Controls	50 ± 8		12 ± 4
2-TPA 150 mg/kg per os	33 ± 5[c]		17 ± 4[d]
Controls	60 ± 9	4.6 ± 2.3	
2-TPA 75 mg/kg per os	41 ± 6[c]	5.8 ± 1.4	
2-TPA 150 mg/kg per os	43 ± 12[d]	7.8 ± 2.9[d]	

[a] Rats were killed 4 hr post-treatment.
[b] Values are the mean ± SD of 8 to 10 observations.
[c] $P < 0.001$ controls vs treated.
[d] $P < 0.05$ controls vs treated.

cemia in a second species, 2-TPA was administered orally to fed guinea pigs at doses of 37.5, 75, and 150 mg/kg. There was no significant effect on blood glucose or liver glycogen during a 4-hr time period.

2-TPTA was administered intravenously at a dose of 75 mg/kg to 4-hr postabsorptive rabbits, again in an attempt to demonstrate hypoglycemic activity in a second species. There was no significant depression in glycemic values at 1, 2, or 4 hr posttreatment. Increasing the dose to 150 mg/kg intravenously resulted in the death of six of nine rabbits.

Overnight-fasted dogs, two dogs per dose group, were orally administered 2-TPTA in gelatin capsules in doses of 37.5, 75, and 150 mg/kg, and jugular vein samples were obtained at various times during a 4-hr period. None of the doses elicited a hypoglycemic effect during the 4-hr period. The effects of the 37.5 mg/kg dose on blood glucose and serum insulin are shown in Fig. 4. At 4 hr post-treatment, both dogs exhibited an increased blood glucose of 14 and 27 mg/100 ml relative to their zero-time value. Serum insulin values had increased 2.5 times the zero-time value 40 min after the administration of the compound. These increases obviously were not associated with a hypoglycemic response.

Discussion. 2-Triphenylphosphoranylideneacetophenone and the *m*-trifluoromethyl analog induce a hypoglycemic response in nonfasted, fasted, and alloxan diabetic rats. This activity has not been demonstrated in the guinea pig or dog following oral administration or in the rabbit following intravenous administration.

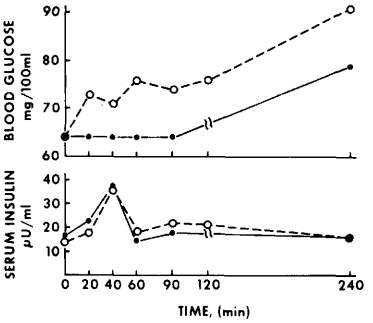


FIG. 4. Effect of 2-TPTA on blood glucose and serum insulin of two fasted dogs. The plots represent the individual values from each dog.

The increase in peripheral serum insulin levels in the rat following the administration of 2-TPA and in the dog following the administration of 2-TPTA suggests that the mechanism in the rat may be due to the stimulation of pancreatic insulin release. Increases in peripheral insulin levels do not reflect the true magnitude of increased pancreatic insulin release, since fasting portal vein insulin levels are twice peripheral vein values (2, 3) with hepatic circulation accounting for a 40–50% extraction of the portal vein concentration (4, 5). In view of these results, increased pancreatic insulin secretion could be substantial. Consistent with the concept of increased insulin release is the slight but significant increase in rat hepatic glycogen, a known physiologic effect of insulin.

Counter to this possible mechanism of action was the ability of both 2-TPA and 2-TPTA to induce a hypoglycemic response in alloxan diabetic rats, an animal model which possesses a relative insulin deficiency due to destruction of pancreatic β cells. Also, the elevated peripheral insulin levels in the dog were not accompanied by a hypoglycemic response.

A number of other 2-triphenylphosphoranylideneacetophenones have been synthesized and tested for hypoglycemic activity (6). Except for the unsubstituted compound 2-TPA, active compounds possessed a meta substituent in the phenyl ring adjacent to the carboxyl carbon. It made no difference whether the meta substituent was an electron-releasing or an electron-withdrawing group. Para or ortho substitution resulted in inactivity. The protonated species, phosphonium salts, of these compounds usually showed activity equivalent to the nonprotonated species.

Summary. The species-specific hypoglycemic activity of two 2-triphenylphosphoranylideneacetophenones is described. 2-Triphenylphosphoranylideneacetophenone (SK&F 45359) and 2-triphenylphosphoranylidene-*m*-trifluoromethyl-acetophenone (SK&F 62775) were hypoglycemic in various rat models, but failed to exhibit hypoglycemic activity in other species.

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