# Comparison of Plasma Insulin Levels Following Administration of Tolbutamide and Glucose

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The mechanism by which sulfonylureas induce hypoglycemia has been a subject of considerable debate. Since the literature in this field is already so vast that it can be touched upon only briefly here, the reader is referred to several excellent reviews.[1-3]

Whereas the bulk of evidence indicates that the hypoglycemic effect in mammals is dependent on the presence of functioning islet β cells and is therefore presumably a consequence of the stimulation of insulin secretion, it has not frequently been possible to demonstrate that the sulfonylureas produce the usual metabolic effects of injected insulin. Notably, studies with $C^{14}$-labeled glucose have generally revealed an unaltered rate of glucose utilization following administration of Na tolbutamide,[4-7] although slight increases were noted in one report.[8] In contrast, insulin administration in similar experiments usually produces a marked increase in $C^{14}$-glucose utilization.[4,9] However, one group of workers[7,10] has found that this effect may be absent if insulin is administered subcutaneously; hypoglycemia in the latter instance presumably results solely from the inhibition of hepatic release of glucose. Such a direct action of insulin on the liver has been disputed by Shoemaker et al.[11] but the objections of the latter group have been overcome by the recent experiments of Madison and co-workers.[12]

It has long been known that the liver is capable of concentrating and destroying insulin. The rapid accumulation of insulin-$I^{131}$ by the liver has been demonstrated in vivo by external counting technics,[13] as well as by direct tissue analysis.[14] It has been shown that the liver removes approximately 50 per cent of insulin-$I^{131}$ administered into the portal vein in a single transhepatic circulation[15] and the studies of Mirsky[16,17] and others[18,19] have demonstrated the marked insulin-degrading capacity of the liver in vitro. Furthermore, since the concentration of endogenous insulin would be higher in portal vein blood than in peripheral blood solely on the basis of dilution, endogenously secreted insulin would be expected to exert a relatively greater hepatic effect and lesser peripheral effect than the same dose of exogenous insulin administered into the peripheral circulation. Jacobs, Reichard, Goodman, Friedmann and Weinhouse[7] have suggested that, in contrast to the effect of exogenous insulin administered into a peripheral vein, the major action of endogenously secreted insulin may be to reduce the output of glucose from the liver rather than to increase glucose utilization by peripheral tissues and that this action could explain the hypoglycemic effect of tolbutamide in the absence of increased peripheral utilization of glucose.

In support of the hypothesis that the absence of definite peripheral effects of sulfonylureas is due to hepatic interception of endogenously secreted insulin are the observations of Pozza, Galansino and Foa,[20] in cross circulation experiments, that pancreatic vein blood but not mesenteric blood shunted into the peripheral circulation of other animals induces hypoglycemia in the recipient animals after tolbutamide administration to the donors.

Concentrations of insulin in the pancreatic vein assayed by the rat diaphragm technic[21] or by means of the hypoglycemic response of intact mice[22] were reported increased following tolbutamide administration but changes were considered of questionable significance when ADH mice were employed for plasma insulin assay.[23] It has also been reported recently that tolbutamide or metahexamide administration to human subjects is followed by marked increases in peripheral blood insulin as assayed with the rat epididymal fat pad.[24]

Since the bio-assays for insulin are of doubtful specificity and subject to the effects of certain ill-defined inhibitory substances reported to be present in plasma and since the difference in effect on peripheral glucose utilization produced by hyperglycemia and sulfonylureas has not yet been satisfactorily explained, it appeared

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of interest to compare changes in the plasma insulin concentration of peripheral blood following glucose and Na tolbutamide administration employing a sensitive and specific immuno-assay for human plasma insulin.[25-27]

## METHODS

Methods employed in the immuno-assay of plasma insulin have been given in detail elsewhere.[26,27] Blood sugar concentrations were determined according to the method of Somogyi[28] or of Folin-Malmros.[29] Twelve fasting nondiabetic male subjects were given 1 gm. Na tolbutamide intravenously and this was followed forty minutes later by 2 mg. glucagon, 25 gm. glucose or 50 gm. glucose intravenously. In three experiments 2 ml. of normal saline was injected twenty minutes prior to the administration of the Na tolbutamide. Blood samples were taken immediately prior to the first injection and at twenty-minute intervals thereafter for a total of five to six specimens. Six nondiabetic subjects and four patients with early maturity-onset diabetes, never treated with insulin, were given 3 gm. Na tolbutamide by mouth after an overnight fast. A standard 100 gm. oral glucose tolerance test was performed under similar conditions within several days preceding or following the tolbutamide study. Blood samples were taken in the fasting state at one half, one and two hours following tolbutamide or glucose administration.

## RESULTS

Blood sugar and plasma insulin concentrations of the four nondiabetic subjects who received intravenous injections of saline followed by Na tolbutamide (1 gm.) and glucagon (2 mg.) are shown in figure 1. Saline had no effect on the plasma insulin levels and tolbutamide produced a rise in all cases but the rise was generally less marked than that following glucagon.

In all patients who received 1 gm. Na tolbutamide intravenously, followed by 25 gm. or 50 gm. glucose intravenously, there was a distinct though temporary increase in insulin concentration following Na tolbutamide administration but this increase was less marked than that observed following glucose administration (figures 2 and 3).

In six nondiabetic and four early maturity-onset diabetic subjects, plasma insulin concentrations rose in every case following 3 gm. Na tolbutamide orally but a much greater increase in plasma insulin was observed following glucose by mouth in the same subjects (figgures 4A and 4B). The mean increase in plasma insulin concentration over the two-hour period was about two and one half to four times as great following glucose as following Na tolbutamide administration in diabetic and nondiabetic subjects (table 1). The blood sugar concentrations during both tests in both groups of subjects are shown in figure 5.

## DISCUSSION

The consistent, albeit modest, increases in immunologically determined plasma insulin following tolbutamide

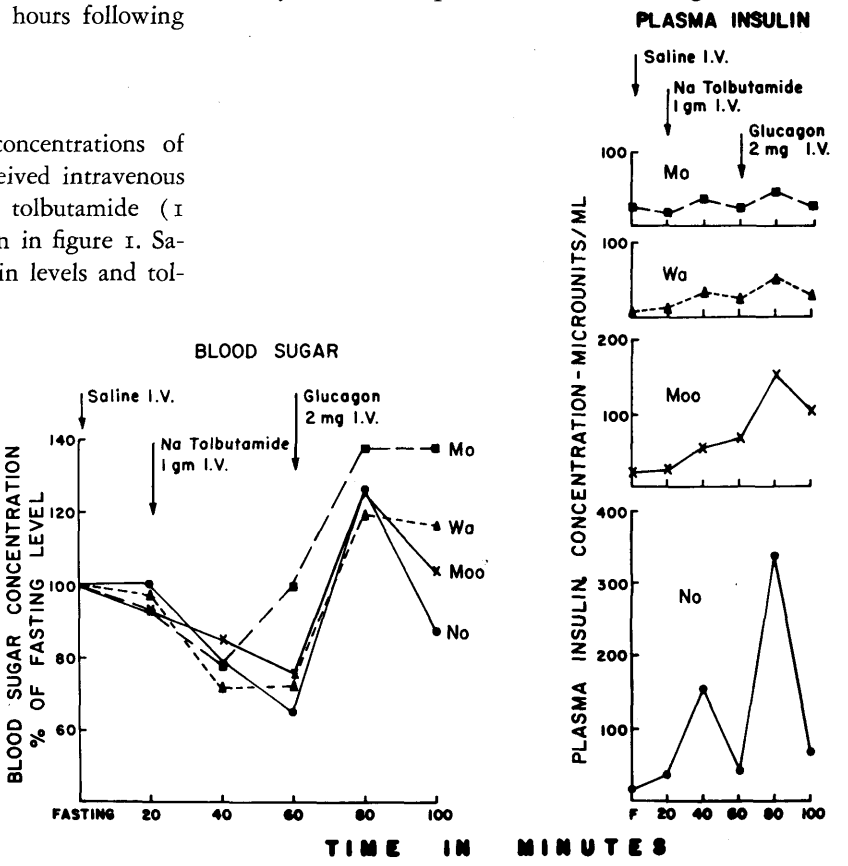


FIG. 1. Blood sugar and plasma insulin concentrations following intravenous administration of saline, 1 gm. Na tolbutamide and 2 mg. glucagon to nondiabetic subjects.

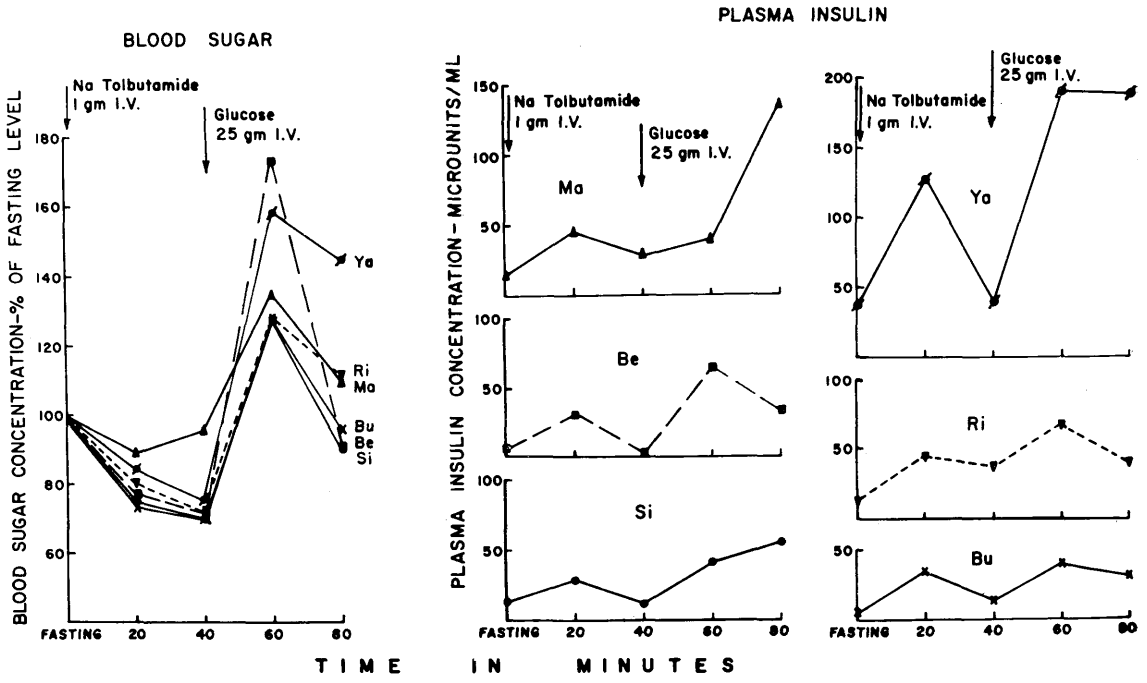


FIG. 2. Blood sugar and plasma insulin concentrations following intravenous administration of 1 gm. Na tolbutamide and 25 gm. glucose to nondiabetic subjects.

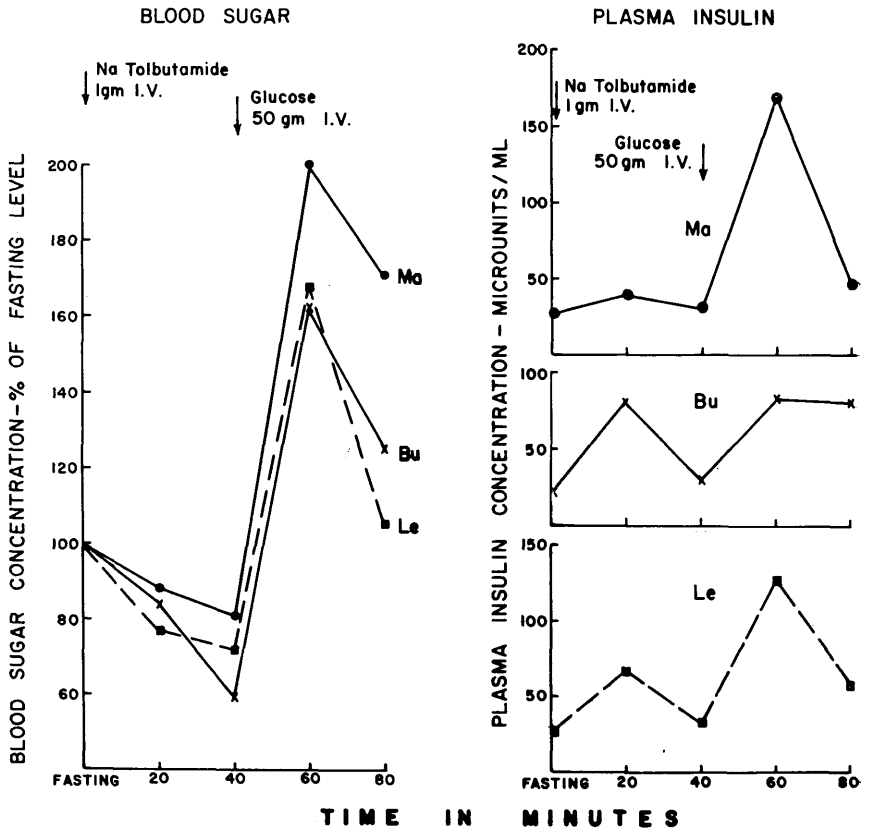


FIG. 3. Blood sugar and plasma insulin concentrations following intravenous administration of 1 gm. Na tolbutamide and 50 gm. glucose to nondiabetic subjects.

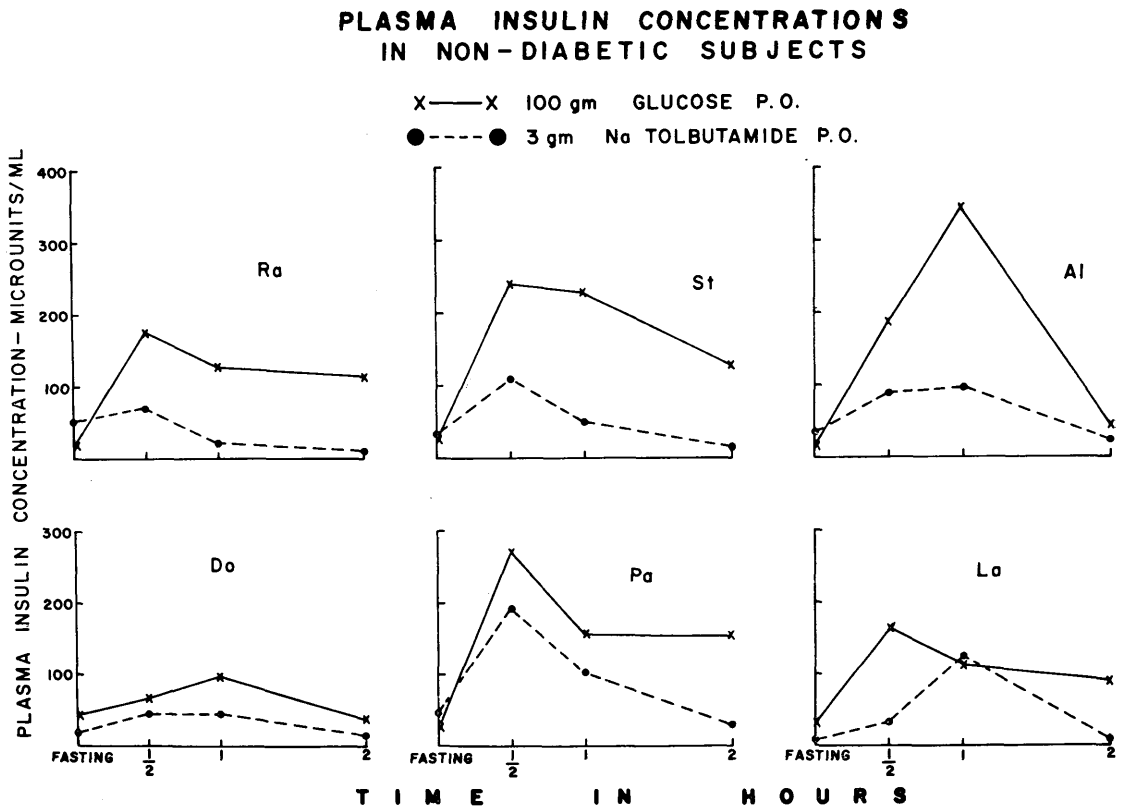


FIG. 4A. Plasma insulin concentrations following oral administration of 3 gm. Na tolbutamide and 100 gm. glucose to fasting nondiabetic subjects.

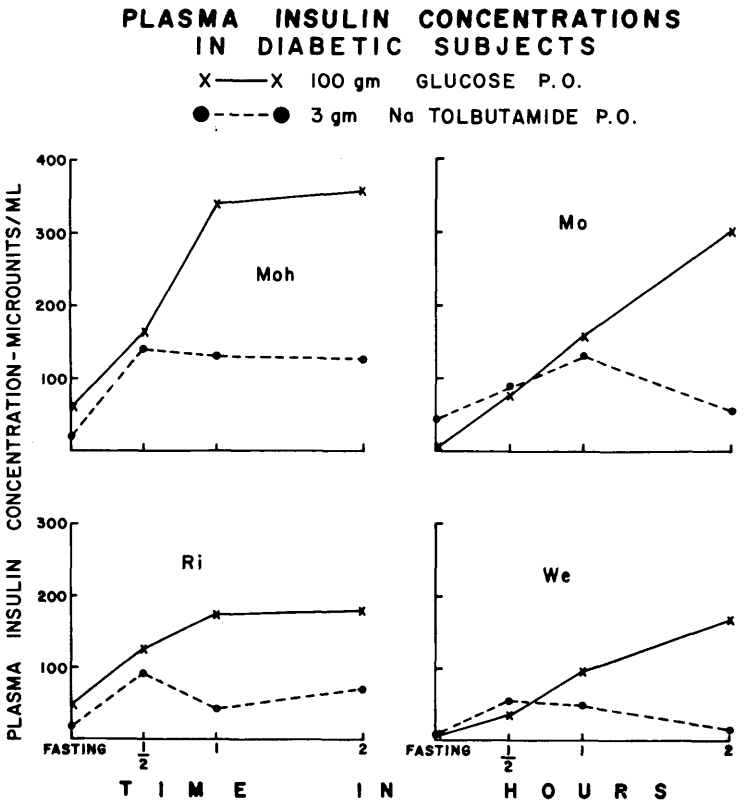


FIG. 4B. Plasma insulin concentrations following oral administration of 3 gm. Na tolbutamide and 100 gm. glucose to fasting diabetic subjects.

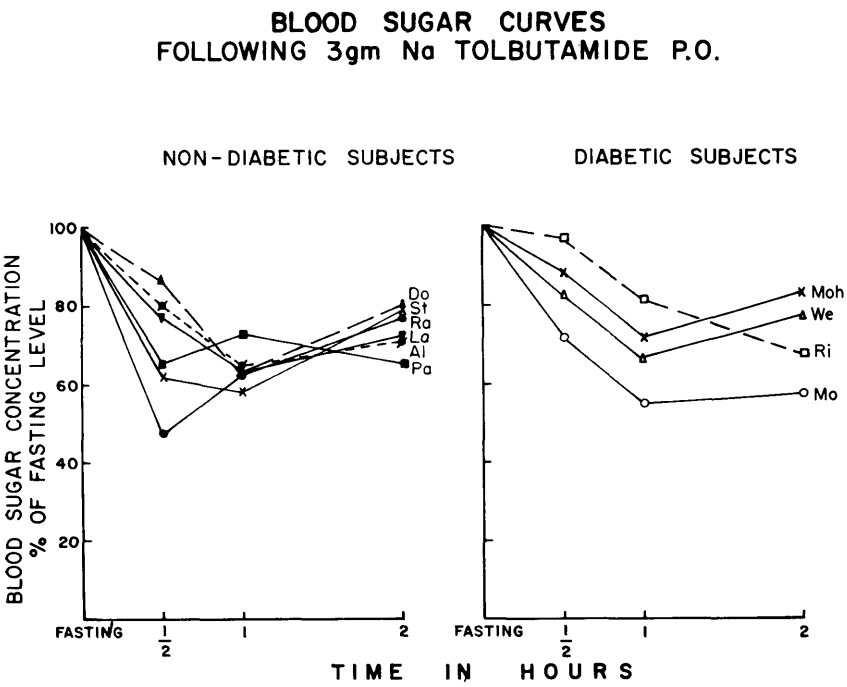


FIG. 5A. Blood sugar concentrations following oral administration of 3 gm. Na tolbutamide to fasting nondiabetic and diabetic subjects.

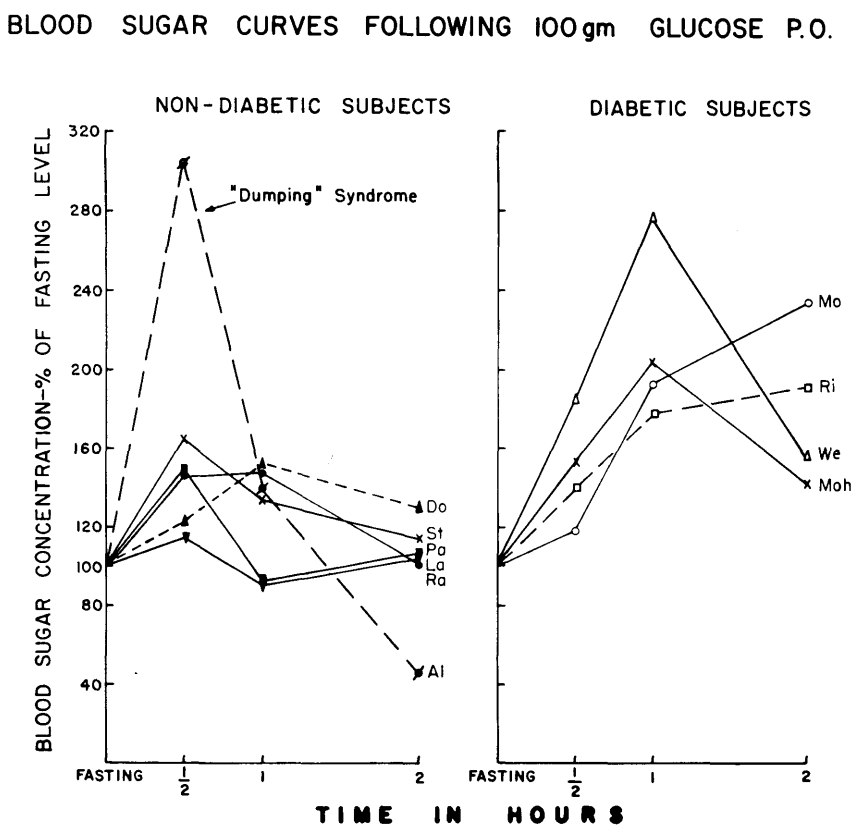


FIG. 5B. Blood sugar concentrations following administration of 100 gm. glucose to fasting nondiabetic and diabetic subjects.

TABLE 1

Mean increase in plasma insulin concentration above fasting level during two-hour period following oral administration of 3 gm. Na tolbutamide or 100 gm. glucose

| Patient | Mean insulin concentration following | |
|---|---|---|
| | Na tolbutamide | Glucose |
| **Nondiabetic subjects** | | |
| Ra | < 0 μU./ml. | 100 μU./ml. |
| Do | 19 | 50 |
| Pa | 50 | 142 |
| St | 18 | 165 |
| Al | 35 | 175 |
| La | 50 | 77 |
| Average | 28 | 119 |
| **Diabetic subjects** | | |
| Ri | 40 | 100 |
| We | 30 | 81 |
| Moh | 92 | 200 |
| Mo | 45 | 147 |
| Average | 52 | 132 |

must be attributed to augmentation of insulin secretion, since the rise in insulin concentration appeared to be too rapid to be explained solely on the basis of inhibited degradation of insulin. Other studies have failed to establish significant effects of hypoglycemic doses of tolbutamide on the rate of insulin-$I^{131}$ degradation in vivo.[30]

Although all patients studied showed increased plasma insulin concentrations following tolbutamide intravenously or orally, the responses usually were significantly less than following administration of glucose by the same route to the same patients. In general, the tolbutamide-induced rise in plasma insulin was of sufficiently moderate degree to be consistent with the absence of detectable effects on peripheral glucose utilization; in some cases the peak insulin concentration following tolbutamide was not higher than is occasionally observed in untreated fasting subjects.[26,27]

The responses in peripheral blood insulin concentration to glucose and tolbutamide administration serve to resolve the different effects of these agents on peripheral glucose utilization. The small increase in insulin secretion stimulated by tolbutamide, while adequate for an action on the liver, is evidently insufficient to produce a demonstrable acceleration of peripheral glucose utilization in the majority of cases, although such an effect might occasionally be exhibited.[8] These observations are consistent with the interpretation of Jacobs et al.,[7] Madison et al.,[12] Craig et al.[31] and others that inhibition of glucose release from the liver in the absence of any peripheral effect on glucose metabolism may result from a small endogenous secretion of insulin stimulated by tolbutamide. This mode of action is compatible with virtually all experimental data on the effects of tolbutamide. However, this interpretation does not necessarily imply that, *in general,* the physiologic effects of endogenously secreted insulin are manifested primarily on hepatic release of glucose. On the contrary, the significantly higher plasma insulin concentrations following glucose administration reported here are consistent with evidence derived from glucose-$C^{14}$ experiments[32,33] that restoration of blood sugar to normal levels following glucose or feeding is effected primarily by increased peripheral utilization of glucose.

## SUMMARY AND CONCLUSIONS

The concentration of insulin in peripheral blood, determined by immuno-assay, was increased in every instance following intravenous administration of 1 gm. sodium tolbutamide in twelve nondiabetic subjects and following oral administration of 3 gm. sodium tolbutamide in six nondiabetic and four early maturity-onset diabetic subjects. However, plasma insulin concentration was increased to a significantly greater extent following glucose administration by the same routes to the same subjects.

These findings are discussed in relation to previous work from which it is concluded that the present results are consistent with the hypotheses: (a) that inhibition of glucose release from the liver in the absence of accelerated peripheral utilization of glucose may result from tolbutamide stimulation of insulin secretion, and (b) that the stimulation of insulin secretion induced by glucose administration results in an increased peripheral utilization of glucose.

## SUMMARIO IN INTERLINGUA

*Comparation del Nivellos de Insulina in le Plasma post Administratationes de Tolbutamida e de Glucosa*

Le concentration de insulina in le sanguine peripheric, determinate per immunoessayage, esseva augmentate sin exception post administrationes intravenose de 1 g de tolbutamida de natrium in dece-duo subjectos nondiabetic e post administrationes oral de 3 g de tolbutamida de natrium in sex subjectos nondiabetic e in quatro subjectos con diabete a declaration in juvene maturitate. Tamen, le concentration plasmatic de insulina esseva augmentate a grados significativemente plus marcate post administrationes de glucosa al mesme subjectos e per le mesme via.

Iste constatationes es discutite in relation a previe investigationes que justifica le assertion que le presente resultatos se trova in congruentia con le hypotheses (a)

que le inhibition del liberation de glucosa ab le hepate in le absentia de un accelerate utilisation peripheric de glucosa pote resultar ab le stimulation del secretion de insulina per le action de tolbutamida e (b) que le stimulation del secretion de insulina inducite per le administration de glucosa resulta in un augmento del utilisation peripheric de glucosa.

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