Effects of taurine on the insulin secretion of rat fetal islets from dams fed a low-protein diet

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Abstract

An isocaloric low-protein (LP) diet (8% instead of 20% in controls) given to dams during gestation reduces the fractional insulin release of stimulated fetal islets. The LP diet lowers the plasma concentration of taurine in both pregnant rats and their fetuses. This study reports the effect of taurine on the *in vitro* release of insulin from control and LP fetal islets. Direct stimulation with taurine, methionine or leucine increased the release of insulin from control islets. Nevertheless, no effect on LP islets was observed with either taurine or methionine. The release of insulin from LP islets was reduced with leucine. The *in vitro* addition of taurine (0.3 or 3 mM) to the culture medium increased the release of insulin from the control islets in response to arginine or leucine, but it did not restore the

reduced responsiveness of LP islets to these amino acids. When 2.5% taurine was added to the drinking water of control or LP dams (groups C+T and LP+T) throughout gestation, the concentration of taurine increased in the serum of dams and fetuses of both groups. The release of insulin from the LP+T fetuses was restored to control levels when stimulated with taurine, methionine, leucine or arginine. In conclusion, taurine stimulated control fetal islets *in vitro*, but failed to do so in LP islets. However, the addition of taurine to the diet of LP dams restored to normal the release of insulin from LP fetal islets, indicating the importance of taurine during development for a normal fetal β cell function.

Journal of Endocrinology (1998) **159**, 341–348

Introduction

Taurine (2-amino ethanesulfonic acid), a sulfur-containing amino acid, is found in almost all mammalian tissues and constitutes more than 50% of free amino acids in many of these tissues (Jacobsen & Smith 1968). Although a considerable amount of this amino acid is also present in the pancreas (Briel *et al.* 1972), its physiological role in this organ is not fully defined. Several reports indicate a possible role of taurine in the regulation of glucose metabolism. In adult rats, taurine increases glucose uptake by the liver. It enhances glycogenesis, glycolysis, and glucose oxidation in this organ (Kulakowski & Maturo 1984, Huxtable 1992). A single administration of taurine not only decreases the blood concentration of glucose, but also increases the hepatic glycogen content (Dokshina *et al.* 1976). Taurine is also known to have an antidiabetic action, potentiating the secretion of insulin and its hypoglycemic effect (Dokchina *et al.* 1976, Tokunagua *et al.* 1983). In the heart, taurine stimulates glucose utilization (Lampson *et al.* 1983). This amino acid inhibits the increase in serum glucose and insulin that occurs after administration of glucose in Wistar-Kyoto rats (Kulakowski & Maturo 1984). These effects of taurine

might be due to its binding to the insulin receptor (Maturo & Kulakowski 1988).

In fetal rats, we have recently demonstrated that taurine directly stimulates insulin release by fetal islets cultured during 7 days. We also showed that, when taurine is added to the culture medium, it enhances the insulin secretion by fetal islets in response to conventional secretagogues (Cherif *et al.* 1996).

The importance of adequate amount of taurine for normal cell development and function is shown by the numerous pathological consequences that occur when the transport of taurine into the cells is curtailed, either by the lack of plasma taurine resulting from dietary deficiency or by the use of transport antagonists. Several studies have demonstrated a correlation between taurine deficiency and abnormalities such as growth and development defects (Sturman 1993), blindness (Hayes *et al.* 1975, Lake & Malik 1987), cardiac dysfunction (Lake 1993, Eley *et al.* 1994), and immunological insufficiency (Lake *et al.* 1992, Schuller-Levis *et al.* 1990).

Taurine concentrations are lower in the serum of both dams and their fetuses at 21.5 days of gestation, when a low (8%) protein (LP) diet instead of the 20% protein control diet has been given throughout gestation (Reusens

et al. 1995). The *in vitro* insulin secretion is decreased when the LP fetal islets are exposed to arginine and leucine (Dahri *et al.* 1991).

In view of the scarcity of data about the role that reduced plasma taurine resulting from a protein-restricted diet (LP diet) has in the decreased secretion of insulin by LP fetal islets, this study was undertaken in order to investigate (a) the secretion of insulin by LP fetal islets in response to taurine stimulation, (b) if the sensitivity of LP islets to secretagogues can be improved by adding taurine to the culture medium, and (c) whether the specific addition of taurine to the mother's LP diet during gestation restores the *in vitro* insulin secretion of fetal islets.

Materials and Methods

Animals and diets

Virgin female Wistar rats were caged overnight with males and copulation was verified in the morning by inspection of vaginal smears. The animals were housed singly in animal facilities maintained at 25 °C with 12 h light : 12 h darkness cycles and were allowed free access to water. In the first and second series of experiments, one group of rats was fed with a control diet (20% protein) and a second group received an isocaloric LP diet (8% protein) compensated with carbohydrates from the first day until day 21·5 of gestation. Diets were purchased from Hope Farms (Woerden, Holland). The composition of the diet has been described previously (Snoeck *et al.* 1990).

In a third series of experiments, animals were also fed with a control or an isocaloric LP diet, but 50% of the control group and LP group females were supplied with taurine-enriched (2·5% w/v) drinking water throughout the pregnancy. Two more groups were thus created: 20% protein diet with added taurine (C+T) and 8% protein diet with added taurine (LP+T).

Tissue culture media

All cultures, including the digestion of the pancreatic tissue, were carried out in RPMI 1640 medium (Gibco, Grand Island, NY, USA) supplemented with 10% v/v heat-inactivated fetal bovine serum and antibiotics (penicillin 200 U/ml, streptomycin 0·2 mg/ml). The final glucose concentration in the culture medium was 11 mM. The DME/F12 defined medium (1 : 1, v/v, Gibco, Paisley, Scotland), used during the last 2 days of culture, was supplemented with antibiotics (penicillin 200 U/ml, streptomycin 0·2 mg/ml) and with taurine (0 mM (control), 0·3 mM or 3 mM respectively). The final concentration of glucose was 8·6 mM.

Tissue culture and islet incubation

The culture technique described by Mourmeaux *et al.* (1985) was slightly modified for this study. Pancreata of

21·5-day-old fetuses from mothers fed either a control or an LP diet were removed separately, minced and digested with collagenase (Boehringer, Mannheim, Germany). Digestion was stopped by adding cold medium. The tissue digest was washed twice. Tissue samples were suspended in 20 ml medium and gently stirred at room temperature for 60 min. The digests were centrifuged, and pellets resuspended at a ratio of one pancreas : 2 ml of RPMI 1640 medium. Two milliliters of this suspension were finally distributed into 35-mm Petri dishes (Falcon 3001; Falcon plastics, Los Angeles, CA, USA). Culture dishes were incubated for up to 7 days at 37 °C, in a humidified atmosphere of 5% CO₂ in the air, the culture medium being changed daily after the second day.

After the culture period, batches of ten free-floating neoformed islets were picked up and incubated for 2 h at 37 °C in 1 ml Krebs Ringer medium (containing 120 mM NaCl, 5 mM KCl, 2 mM CaCl₂, 1 mM MgCl₂, 22 mM NaHCO₃). The solution was supplemented with 5 mg/ml bovine serum albumin (Fraction V, Calbiochem-Behring, San Diego, CA, USA) and gassed with O₂/CO₂ (95%/5%) to maintain a pH of 7·4. The concentration of glucose was adjusted as required (5·6 or 16·7 mM) and test substances were added without correction for osmolarity. Taurine, leucine or arginine was added in a concentration of 10 mM and methionine at 5 mM, in accordance with their relative concentrations in the fetal rat serum.

At the end of the incubation, the medium was removed and placed in a watch glass in order to verify that no islet had been included, and then frozen before further use in the insulin assay. To determine the insulin content of islets, they were sonicated in 0·5 ml acid-ethanol (0·15 M HCl in 75% (v/v) ethanol in water). In order to obviate variations arising from differences in individual islet batches, insulin secretion during incubation was expressed as a percentage of the islet content of insulin at the start of the incubation. This value was obtained by adding the content measured at the end of the incubation to the amount of released insulin.

In a first series of experiments, in order to study the acute effect of taurine on insulin secreted by the LP group and control group fetal islets in subsequent incubations, RPMI 1640 medium was used during the entire culture period (7 days). The islets were incubated as described in the presence of glucose (5·6 mM) with or without taurine (10 mM), leucine (10 mM) or methionine (5 mM).

To investigate the effect of a chronic supply of taurine on the secretion of insulin by fetal LP and control group islets, culture dishes were washed twice with DME/F12 medium after 5 days of culture, and cultured for 2 more days in DME/F12 medium without or with 0·3 or 3 mM taurine. Neoformed islets were then incubated in the presence of glucose alone (5·6 or 16·7 mM) or glucose (5·6 mM) with leucine (10 mM) or arginine (10 mM).

Finally, pancreata of 21·5-day-old fetuses from dams fed either a control, a C+T, a LP or a LP+T diet were

cultured during 7 days in the presence of RPMI 1640 medium. This allowed us to study how taurine-supplemented diets fed to the mother during pregnancy affected the *in vitro* secretion of insulin by fetal islets in response to secretagogues. Islets were incubated in the presence of glucose alone (5.6 or 16.7 mM) or glucose (5.6 mM) added to taurine, leucine, arginine (each 10 mM) or methionine (5 mM).

Test substances

D-Glucose, L-leucine and L-arginine were provided from Merck (Darmstadt, Germany). L-Taurine and L-methionine were provided from Sigma (St Louis, MO, USA).

Insulin assay

The amounts of insulin released and the insulin content were measured with a radioimmunoassay kit (Insik 5 p2796; Sorin, Italy). Rat insulin (Novo-Nordisk, Denmark) was used to establish a standard curve, using immunoreactive insulin (IRI) guinea pig-antihuman insulin antiserum and porcine monoiodinated ^{125}I -insulin. The binding complex was precipitated with an antibody raised against guineapig IgG. Samples were then centrifuged at 1500 *g* in order to separate free from bound hormone. The method allowed the determination of 4 $\mu\text{M}/\text{ml}$ (0.16 ng/ml) with variation coefficients within and between assays of 8.2%.

Measurement of taurine plasma concentrations

At 21.5 days of gestation, all rats were anesthetized with sodium pentobarbital (55 mg/kg body weight). Fetal blood samples were rapidly collected through axillary vessels, the feto-maternal circulation being maintained. Maternal blood was also collected in heparinized vials. The measurement of taurine was performed by a HPLC procedure as described by Donzanti & Yamamoto (1996). All reagents used were analytical grade, except for HPLC-grade methanol, and purchased from Sigma. Plasma samples were deproteinized, filtered through 0.2- μm cellulose nitrate filter (Gelman Sciences, Ann Arbor, MI, USA) and derivatized with *o*-phthalaldehyde (OPA). For this purpose, 27 mg OPA were dissolved in 1 ml methanol contained in 10 ml β -mercaptoethanol (BME). This solution was diluted with 9 ml 0.1 M sodium tetraborate buffer pH 9.3 and stored at 4 °C. The working solution (OPA/BME) was prepared 24 h before use by diluting 1 ml of the above solution in 3 ml 0.1 M sodium tetraborate. The derivatization procedure entailed mixing the sample and standard with 10 μl OPA/BME for 2 min in complete darkness before injection into the HPLC system (Model 7125 Rheodyne, Berkeley, CA, USA; LDC Cousta Metric 3200 pump). Twenty-microliter samples were separated on a Biophase-II column

Figure 1 Direct effect of taurine, methionine or leucine on fractional insulin release of control (C) or LP fetal islets. Islets were cultured in a medium containing 8.2 mM glucose. After culture, islets were stimulated for 2 h with glucose alone (G 5.6 mM) or with glucose added to taurine (Tau 10 mM), methionine (Met 5 mM) or leucine (Leu 10 mM). Values are the means of 12 observations pooled from six independent cultures ($n=12$). Bars represent S.E.M. ^a $P<0.01$ compared with each control; ^b $P<0.01$ compared with glucose stimulation.

(100 $\times$ 3.2 mm, octadecyl sulfate 3 μm) and eluted with 32% methanol in 0.13 mM EDTA/0.1 M NaHPO_4 buffer at pH 6.4 at a flow rate of 1 ml/min. Samples were then detected coulometrically (ESA, Inc., Belford, MA, USA) using three electrodes: a guard (0.4 V), preoxidation (-0.4 V) and working (+0.6 V) electrode (Analytical cell ESA Model 5011).

A PC integration Pack (Kontron Instruments) was used to quantify taurine with a standard solution containing 1 mM taurine in 50% v/v methanol.

Statistics

Statistical comparisons were made using one- or two-way analysis of variance (ANOVA) followed by a Scheffé's test. However, for the analysis of taurine concentrations in the serum of dams and fetuses, statistical analysis was performed on log-transformed values because of the heterogeneity of variances. Then, analysis of variance against two fixed and crossed classification criteria (ANOVA 2), followed by a Tukey test was used.

Results

Insulin secretion and content of fetal islets stimulated with taurine

Neoformed control or LP islets cultured during 7 days in RPMI 1640 medium were incubated in the presence of glucose alone (5.6 mM), or supplemented with leucine (10 mM), taurine (10 mM) or methionine (5 mM). In the control group, taurine produced an increase in insulin release similar to that seen with leucine or methionine ($P<0.01$; Fig. 1), whereas, in the LP group, neither

Table 1 Effect of secretagogues on the insulin content of islets from control (C) or LP fetuses before the incubation. Islets were cultured for 7 days in RPMI 1640 medium, and incubated for 2 h in Krebs Ringer solution supplemented with glucose (G) alone (5.6 mM) or with added leucine (Leu), taurine (Tau) (each at 10 mM) or methionine (Met) (5 mM). Values are means ± S.E.M. (n=6)

Secretagogue (mM)	Insulin content (ng/islet)	
	C	LP
G 5.6	34.2 ± 5.0	35.6 ± 2.7
G 5.6+Leu 10	29.7 ± 2.3	31.4 ± 0.8
G 5.6+Tau 10	34.5 ± 2.6	33.4 ± 3.0
G 5.6+Met 5	38.0 ± 3.6	32.1 ± 1.6

taurine nor methionine had any effect on insulin secretion. Leucine stimulated insulin secretion by LP fetal islets, but this response was 50% lower than that of control islets. The response of LP islets to glucose alone (5.6 mM) was also diminished by about 50% compared with that in the controls. Two-way ANOVA showed a significant difference in release of insulin between control and LP groups ($P<0.01$) in response to all secretagogues investigated; however, the measurement of the insulin content of the islets did not reveal any difference between control and LP groups (Table 1).

Insulin secretion and content of fetal islets cultured with taurine

The islets were cultured in a medium containing 8.2 mM glucose, to which 0.3 or 3 mM taurine were added for the last 2 days. Islets were then incubated in a Krebs Ringer solution, in the presence of glucose (5.6 or 16.7 mM) alone or glucose (5.6 mM) with leucine or arginine (each at 10 mM).

Control fetal islets failed to respond to glucose, but their insulin secretion increased in response to leucine and arginine. The presence of taurine in the culture medium enhanced the response of islets to the secretagogues in the subsequent incubation (Fig. 2a). More specifically, both low (0.3 mM) and high (3 mM) taurine concentrations enhanced insulin release when islets were stimulated with glucose alone (5.6 or 16.7 mM, $P<0.05$; $P<0.01$ respectively). When islets were stimulated with leucine or arginine in the presence of glucose (5.6 mM), 3 mM taurine concentration was necessary to produce a significant increase in the insulin response ($P<0.01$). ANOVA 2 did not reveal any significant difference in the insulin content between islets grown in the absence or in the presence of taurine (0.3 or 3 mM) (Table 2).

In islets of the LP group cultured in the absence of taurine, both leucine and arginine enhanced insulin secretion, but glucose had no effect (Fig. 2b). Insulin secretion of LP fetal islets was not affected by the presence

Figure 2 Effect of taurine in the culture medium on fractional insulin secretion of (a) fetal control islets (C) and (b) LP islets. Islets were cultured in a medium containing 8.2 mM glucose without (0 mM) or with taurine (0.3 or 3 mM). After the culture, islets were incubated for 2 h in presence of glucose (G) alone (5.6 or 16.7 mM) or added to leucine (Leu) or arginine (Arg) (each at 10 mM). Values are the means of nine observations pooled from three independent cultures (n=9). Bars represent S.E.M. ^a $P<0.05$, ^b $P<0.01$ compared with each internal control (taurine 0 mM), ^c $P<0.01$ compared with glucose stimulation in the taurine 0 mM group.

of taurine in the culture medium, and there was no difference in insulin content between islets grown in the absence of taurine and those grown in the presence of

Table 2 Effect of a chronic supply of taurine on the insulin content of islets from control (C) or LP fetuses before the incubation. Islets were cultured for 5 days in RPMI 1640 medium, and for 2 days in a defined medium DME/F12 in the presence of 8.6 mM glucose without (0 mM) or with 0.3 or 3 mM taurine. Neoformed islets were then incubated in Krebs Ringer solution supplemented with glucose alone (5.6 or 16.7 mM) or with glucose (5.6 mM) added to leucine or arginine (each at 10 mM). Values are means ± S.E.M. (n=24)

Taurine concentration (mM)	Insulin content (ng/islet)	
	C	LP
0	31.0 ± 1.8	32.0 ± 2.6
0.3	29.7 ± 3.5	33.3 ± 1.5
3	30.0 ± 1.7	31.8 ± 3.4

0.3 mM or 3 mM taurine (Table 2). The insulin content of control and LP fetal islets was similar in each experimental condition.

Insulin secretion and content of control and LP fetal islets from dams treated with taurine

Fetal and maternal blood samples were collected. Taurine concentration was measured and found to be reduced in the serum of LP dams and their fetuses when compared with controls ($P<0.05$; Fig. 3a, b respectively). Addition of taurine to the drinking water of control and LP dams increased the concentration of this sulfur amino acid in the plasma of mother and fetus in both groups ($P<0.05$).

After 7 days of culture, fetal islets from the four groups (control, C+T, LP and LP+T) were incubated in the presence of glucose alone (5.6 or 16.7 mM) or glucose (5.6 mM) supplemented with taurine, arginine, leucine (each at 10 mM) or methionine (5 mM). Figure 4 shows that, in the control group, fetal islets did not respond to glucose, but their insulin secretion increased in response to amino acids ($P<0.01$). When taurine was added to the mother's diet, insulin secretion of C+T fetal islets significantly increased in response to glucose (16.7 mM; $P<0.01$), and methionine ($P<0.01$) compared with controls in each stimulatory condition. Compared with the control group, insulin secretion of the LP fetal islets was reduced in response to leucine and arginine ($P<0.01$). LP fetal islets did not respond to taurine or methionine stimulations ($P<0.01$ compared with the response in the control group).

When taurine was added to the drinking water of the LP dams (Fig. 4), insulin secretion of LP+T fetal islets was increased in response to glucose (16.7 mM; $P<0.05$), leucine, arginine, taurine ($P<0.01$) and methionine ($P<0.05$) compared with the response of the LP islets. This increase attained the level of insulin response seen in control islets. No difference in the insulin content of islets of fetuses from the four groups was observed when taurine was administered to dams (Table 3).

Discussion

The current study highlights, for the first time, the importance of normal taurine plasma concentrations during intrauterine development for normal fetal secretion of insulin. We have presented new data on insulin secretion by LP fetal islets in response to the stimulation by taurine, after culture in the presence of this sulfur amino acid and when the mother was fed a taurine-supplemented diet during gestation.

In agreement with our previous results, the addition of taurine to the culture medium enhanced the secretion of insulin by normal islets in response to secretagogues. Furthermore, both taurine and methionine directly stimu-

Figure 3 Taurine concentration in the serum of (a) dams fed control (C) or LP diets without taurine or with taurine (C+T; LP+T) and (b) their fetuses. Values are the means of six animals. Bars represent S.E.M. ^a $P<0.05$ LP compared with control and C+T compared with control; ^b $P<0.05$ LP+T compared with LP.

lated insulin secretion to the same extent as did leucine (Cherif *et al.* 1996). However, this direct stimulation by taurine or methionine was not observed with LP islets. Because taurine concentrations were previously found to be more affected than those of other amino acids in the serum of the LP dams and LP fetuses (Reusens *et al.* 1995), it was hypothesized that addition of taurine to the culture medium or to the dam's diet could improve the *in vitro* pattern of insulin secretion of the LP fetal islets. However, the insulin secretion by LP fetal islets in response to leucine or arginine remained reduced even after these islets were cultured in the presence of 0.3 or 3 mM taurine. Taurine in the culture medium was thus unable to restore the sensitivity of islets from the LP group to amino acid secretagogues. Addition of taurine to the mother's

Figure 4 Fractional release of insulin from fetal islets in relation to maternal isocaloric diet containing 20% or 8% protein, each without or with a dietary supplement of taurine. Fetal islets were cultured for 7 days in a medium containing 8.2 mM glucose. Islets were then stimulated for 2 h with glucose (G) alone (5.6 or 16.7 mM) or with glucose (G 5.6 mM) and taurine (Tau 10 mM), methionine (Met 5 mM), leucine (Leu 10 mM) or arginine (Arg 10 mM). Values are the means of eight observations pooled from three cultures independently ($n=8$). Bars represent S.E.M. ^a $P<0.01$ LP compared with control in each stimulatory condition; ^b $P<0.01$ C+T compared with control in each stimulatory condition; ^c $P<0.05$ LP+T compared with LP in each stimulatory condition. ^d $P<0.01$ LP+T compared with LP in each stimulatory condition.

diet resulted in an increased taurine concentration in plasma of pregnant rats and their fetuses; it did restore to normal the insulin secretion of the LP fetal islets. This effect of *in vivo* supplementation of taurine, which was not observed when taurine was added *in vitro*, could be explained by the fact that LP+T fetal islets have originated, differentiated and proliferated in an environment in which there was an adequate amount of taurine during intrauterine development. In another study, when taurine was given during 11 days to adult rats fed an 8% protein diet from fetal life until adulthood, the secretion of insulin in response to nutrient secretagogues remained lower than that in control rats (fed a 20% protein diet) (Scruel *et al.*

1997). The restoration of taurine plasma concentrations only at adulthood, observed by Scruel *et al.* (1997), was thus ineffective in correcting insulin secretion. These results emphasize the need for adequate plasma concentrations of taurine at least during fetal development, in order to have normal fetal insulin secretion. The current investigation demonstrates that taurine is involved specifically in the development of the insulin secretory process at the fetal stage.

The difference between the release of insulin by control and LP islets, which disappeared only when taurine was added *in vivo*, could be due to low expression, low activity, or both, of a taurine transporter in fetal LP islets. The

Table 3 Effect of a taurine-supplemented diet on the insulin content of islets from control (C), LP, C+T and LP+T fetuses at the beginning of the incubation. Islets were cultured for 7 days in RPMI 1640 medium, and incubated for 2 h in Krebs Ringer solution supplemented with glucose (G) alone (5.6, 16.7 mM) or G 5.6 mM with added leucine (Leu), arginine (Arg), taurine (Tau) (each at 10 mM) or methionine (Met) (5 mM). Values are means $\pm$ S.E.M. ($n=8$)

Secretagogue (mM)	Insulin content (ng/islet)			
	C	C+T	LP	LP+T
G 5.6	22.1 $\pm$ 1.7	28.0 $\pm$ 4.1	26.6 $\pm$ 4.6	34.9 $\pm$ 2.5
G 16.7	29.1 $\pm$ 4.6	23.0 $\pm$ 1.3	24.1 $\pm$ 2.8	32.8 $\pm$ 2.8
G 5.6+Leu 10	24.6 $\pm$ 2.5	23.6 $\pm$ 2.3	27.1 $\pm$ 3.7	34.5 $\pm$ 5.1
G 5.6+Arg 10	29.0 $\pm$ 3.2	22.4 $\pm$ 1.8	26.0 $\pm$ 1.4	29.0 $\pm$ 3.0
G 5.6+Tau 10	28.9 $\pm$ 2.8	28.2 $\pm$ 2.9	28.2 $\pm$ 4.2	28.4 $\pm$ 3.4
G 5.6+Met 15	27.1 $\pm$ 3.8	27.0 $\pm$ 1.6	30.4 $\pm$ 4.2	30.0 $\pm$ 1.8

pancreas is known to contain taurine (Briel *et al.* 1972), but its taurine transporter has not been identified, although such transporters in the brain (Smith *et al.* 1992), placenta (Ramamoorthy *et al.* 1993), kidney and retina (Leibach *et al.* 1993) and thyroid cells (Jhiang *et al.* 1993) have been cloned and characterized.

The difference in release of insulin by control and LP islets could also be related to changes in Ca^{2+} stores or movements known to affect insulin secretion mechanisms (Grodsky & Bennett 1966, Wolheim *et al.* 1975, Gilon *et al.* 1993). The low taurine concentration known to occur in the plasma of our experimental LP group could have influenced the Ca^{2+} pattern of islets during development, as taurine modulates many Ca^{2+} -dependent processes (Huxtable 1992, Porter & Martin 1993). The cellular Ca^{2+} stores in mitochondria and endoplasmic reticulum are known to be regulated by taurine (Lombardini 1988), which also controls the activity of two transport systems, present in the plasma membrane of cells, that are responsible for the efflux of Ca^{2+} from the cell: the $\text{Ca}^{2+}/\text{Mg}^{2+}$ -ATPase and the Na^{+} - Ca^{2+} exchanger (Igisu *et al.* 1976). Investigations are in process to determine the effect of taurine on calcium movements in fetal B cells.

In addition, the current study has confirmed previous data showing that glucose was unable significantly to stimulate release of insulin from control fetal islets (Weinhaus *et al.* 1995, Tu & Tuch 1996). A possible explanation for this immaturity of function is an insufficient glucose oxidative phosphorylation in mitochondria with inadequate synthesis of ATP (Hughes 1994) and a failure to close ATP-dependent K^{+} channels (Rorsman *et al.* 1989). This immaturity might also be due to reduced glucokinase activity (Tiedge & Lenzen 1993). However, Tu & Tuch (1997) do not support this latter hypothesis, but favour that of Hughes (1994).

In conclusion, during gestation, supplementation of taurine in the LP diet of dams did restore to normal the *in vitro* reduced insulin secretion of fetal LP islets. However, addition of taurine to the culture medium failed to normalize the insulin secretion of fetal LP islets that developed in the presence of a low plasma concentration of taurine. We suggest, therefore, that an adequate amount of taurine is specifically needed during development for the normal function of fetal B cells.

Acknowledgements

We gratefully acknowledge Professor P De Witte and Dr A Dahchour for their help in taurine measurement and Professor E Le Boulenger for his advice in statistical analysis.

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Received 11 September 1997

Accepted 13 July 1998