J. Pharm. Pharmacol. 1997, 49: 873–877
Received January 27, 1997
Accepted May 14, 1997

Effects of Trecadrine, a β_3-Adrenergic Agonist, on Intestinal Absorption of D-Galactose and Disaccharidase Activities in Three Physiopathological Models

ANA DIEZ-SAMPEDRO, FERMIN I. MILAGRO, BEATRIZ BERRAONDO, MARIA A. ZULET, J. ALFREDO MARTINEZ AND ANA BARBER

Department of Physiology and Nutrition, University of Navarra, c/Irunlarrea s/n, Pamplona 31008, Spain

Abstract

Impairments in intestinal absorptive and digestive processes have been described in several pathophysiological situations, such as in drug-induced diabetes, obesity and hypercholesterolaemia. Furthermore, there is evidence for the occurrence of β_3-adrenoceptors in multiple regions of the gastrointestinal tract, but there are no data concerning their possible involvement on jejunal and ileal digestive and absorptive functions. In this work, we have measured the modifications of selective intestinal absorption and disaccharidase activities in alloxan-induced diabetic and in diet-induced obese and hypercholesterolaemic Wistar rats. The action of a β_3-adrenergic agonist (Trecadrine) with hypoglycaemic and lipolytic properties on those gastrointestinal functions has been studied.

Increases in the galactose uptake by intestinal rings and in both sucrase and maltase activities were found in diabetic rats. The results obtained after Trecadrine administration to diabetic rats led to an improvement of the altered values. On the other hand, our data show a decrease in sugar absorption and in disaccharidase activities in both obese and hypercholesterolaemic groups, probably related to the low carbohydrate and high fat content of these diets. An amelioration in sucrase activity was observed after treatment with Trecadrine. Finally, Trecadrine administration to control animals significantly inhibited galactose intestinal absorption, which was independently confirmed by additional in-vitro studies.

Overall, these results could be attributed not only to an improvement in the pathophysiological condition (diabetes, obesity and hypercholesterolaemia), but also to a direct effect of the β_3-adrenergic agonist on the intestinal absorption processes.

Changes in intestinal structure and function have been observed in different animal models associated with a great variety of factors, including hyperglycaemia (Nashiro et al 1992) and high-carbohydrate (Ferraris et al 1993) or high-fat (Goda & Takase 1994) intake. In this way, variations in jejunal transport of monosaccharides and in disaccharidase activities have been found in several pathophysiological situations, such as in drug-induced diabetes (Nakabou et al 1985), and in both genetically (Ferraris & Vinnakota 1995) and diet-induced (Planas et al 1992) obese rodents.

A number of drugs have been reported to play a role in digestive and absorptive processes, by modifying the transport of monosaccharides or the activity of disaccharidases, or both. Thus, some antibiotics, such as cephalosporins (Idoate et al 1996), and different α_1- (McIntyre et al 1992), α_2- (Barry et al 1993) and β- (McIntyre et al 1993) adrenergic agonists, may influence some gastrointestinal activities.

Recently a new subgroup of β-adrenergic agonists (β_3) with potential antidiabetic and anti-obesity applications has been described (Howe 1993). There is some evidence, from functional and binding studies, for the presence of β_3-adrenoceptors in a number of tissues, including brown and white adipose tissue, skeletal muscle, brain, gall bladder and multiple regions from the gastrointestinal tract, such as in the oesophagus, gastric fundus, jejunum, ileum and colon (reviewed by Manara et al 1995), being more numerous in smooth muscle and in submucosa than in mucosa itself (Evans et al 1996). Recently it has been proved that these β_3-adrenoceptors can play an important role in the regulation of the intestinal motility by disrupting the migrating myoelectric complex pattern (Thollander et al 1996). However, at present there are no data about their possible involvement in jejunal and ileal digestive and absorptive functions.

In the present study, we have measured the sugar digestive and absorptive processes in diet-induced obese and hypercholesterolaemic animals as well as in alloxan-induced diabetic Wistar rats. This experimental trial was conducted to determine whether a new orally administered β_3-adrenergic agonist, Trecadrine, a diphenyl-methylen-ethylamine derivative compound with hypoglycaemic and lipolytic properties (Barrionuevo et al 1996), may contribute to a recovery in the digestive and absorptive altered functions. Also we investigated whether these effects may be due to the normalisation of the pathophysiological condition, or may occur through a direct role of β_3-adrenoceptors in the digestive and absorptive processes.

Correspondence: A. Barber, Department of Physiology and Nutrition, University of Navarra, c/Irunlarrea s/n, Pamplona 31008, Spain.

Materials and Methods

Animals

Wistar rats weighing about 200 g, obtained from the Center of Applied Pharmacology (C. I. F. A.), were used for all experimental trials. In all the cases, rats were maintained under regulated environmental conditions with artificial light from 0800 to 2000 h and a temperature of 22 ± 2°C.

Drugs

Trecadrine, used as a pure compound, is a diphenyl-methyl en-ethylamine derivative, whose formula has been previously published (Barrionuevo et al 1996). This product was generously supplied by Wassermann-Chiesi.

Basal model

Firstly, sixteen female Wistar rats were divided into two groups (control group, n = 8, Trecadrine group, n = 8) housed in cages; nutritionally balanced pellets and water were freely available. Rats from the trecadrine group were orally dosed by gavage with Trecadrine (1 mg kg^{-1} day^{-1}) for 35 days, while rats from the control group received a placebo (NaCl 0·9%). After this period, rats were killed by decapitation, and basal determinations were performed in this group, concerning absorptive and digestive processes (see below).

Diabetes model

Eighteen female Wistar rats were housed in individual metabolic cages, given free access to pellets and water, and divided into two groups. Rats from the first group (n = 10) received a single subcutaneous dose of alloxan (150 mg k^{-1}) in 0·9% NaCl to make them diabetics, while rats from the second group (n = 8) received a placebo (NaCl 0·9%). Two days later, some of the diabetic animals (Diabetes $+\beta_3$, n = 5) were given trecadrine orally (1 mg kg^{-1} day^{-1}) for three days, while the other diabetic (diabetes group, n = 5) and control rodents received a placebo. On the fourth day, all rats were killed by decapitation and exsanguinated.

Plasma glucose was measured with a dry-chemistry procedure (Spotchem, Menarini), in order to ascertain whether the diabetic state was reached. Animals were considered diabetic if the glucose levels were greater than 11 mmol L^{-1}.

Obesity model

Twenty-four female Wistar rats were housed in cages in groups of four, and initially divided into two groups. The first group (control group, n = 8) was fed pellets and water freely, while the second (obese group, n = 16) was fed on a cafeteria diet (hypercalorific, high-fat) containing: pate, chips, chocolate, bacon, biscuits and pellets, in a proportion of 4 : 2 : 2 : 2 : 2 : 1 presented in excess, in addition to freely available water. After 40 days, animals fed with the cafeteria diet (obese group, n = 16) became obese, and then were divided into two new groups (obese group, n = 8; obese $+\beta_3$ group, n = 8). Rats from the obese $+\beta_3$ group were dosed orally by gavage with trecadrine (1 mg kg^{-1} day^{-1}) for 35 days, while rats from the control and obese groups received a placebo. After 75 days, rats were decapitated.

Animals were weighed regularly and body composition was measured at the end of the experimental period by using a non-invasive electromagnetic technology (EM-SCAN Model SA-2) after animals were anaesthesized with ether.

Hypercholesterolaemia model

Thirty male Wistar rats were divided into two groups and fed on different dietary formulations (Zulet & Martínez 1995). The first group (control group, n = 10) received a semipurified control diet and the second (hyper group, n = 20) was fed on a diet enriched in saturated fat (25%) and cholesterol (1%). After 26 days, the hypercholesterolaemic state was reached. Moreover, for 15 days all the animals continued on the same diet but ten hypercholesterolaemic animals were also dosed orally with trecadrine (1 mg kg^{-1} day^{-1}) (hyper $+\beta_3$ group, n = 10). Before they were killed by decapitation, serum was separated from blood samples, and total cholesterol and high density lipoproteins (HDL) were measured by using appropriate kits from Boehringer Mannheim (Boehringer Mannheim, France S. A.), to assess degree of hypercholesterolaemia achieved.

Studies on galactose uptake

After the animals were killed by decapitation, the abdomen was opened and jejunum was removed, rinsed with saline solution and everted (Lugea et al 1994). Briefly, the rat jejunum was cut into small rings (about 0·5 cm) and four rings selected randomly were incubated in 10 mL of saline solution containing 2·0 mM D-gal with 0·25 μCi [1-^{14}C]-gal. The composition of the physiological solution was (mM): 140·0 NaCl, 5·6 KCl, 3·0 $CaCl_2$, 2·8 KH_2PO_4, 2·8 $MgSO_4$, 6·1 Tris and 4·9 HCl (pH 7·4). The incubation was carried out for 15 min at 37°C with continuous oxygenation (95% O_2–5% CO_2). At the end of the incubation period, the rings were removed, washed with ice-cold saline solution, weighed and extracted for 24 h in 0·5 mL 0·1 M nitric acid. Samples from both the incubation solution and the tissue extracts were taken and the radioactivity was measured by liquid scintillation counting.

Additionally, to study the in-vitro effect of trecadrine, intestinal rings from male Wistar rats were used. In this case, trecadrine was added directly to the saline solution in a final concentration of 10^{-5} M and the same experimental procedure (Lugea et al 1994) was followed.

Preparation of brush-border membrane vesicles and assay of enzymes

Brush-border membrane vesicles were prepared according to the procedure described by Shirazi-Beechey et al (1990). All steps were carried out at 0–4°C. Briefly, the jejunum of control and treated animals was washed with NaCl 0·9%, everted and resuspended in a buffer containing 100 mM mannitol and 2 mM HEPES-Tris buffer, pH 7·4. The enterocytes were removed by using a Vibromixer (model E-1, Omnimix, Sorvall), $MgCl_2$ was added to the suspension to a final concentration of 10 mM and mixed for 20 min before centrifugation at 2000 *g* for 15 min. The supernatant was centrifuged at 27 000 *g* for 30 min to produce a pellet which was then subjected to further homogenization in a buffer containing 100 mM mannitol, 0·1 mM $MgSO_4 \cdot 7H_2O$ and 2 mM HEPES-Tris buffer, pH 7·4 and centrifugation at 27 000 *g* for 30 min. The final pellet was resuspended in 1 mL solution which contained 300 mM mannitol, 0·1 mM $MgSO_4 \cdot 7H_2O$ and 10 mM HEPES-Tris buffer, pH 7·4. The digestion of disaccharides (maltose, sucrose) was assayed in brush-border vesicles of jejunum by estimating D-glucose formation with the glucose-oxidase-peroxidase method (Dahlqvist 1964). Protein content was determinated by the method of Bradford (1976) with bovine serum albumin as standard.

Statistical methods

All results are expressed as mean ± s.e., and comparisons between means were analysed statistically using the one-way analysis of variance followed by a Fisher PLSD test, or by

Kruskal–Wallis and U–Mann–Whitney tests as appropriate. Differences were considered statistically significant if $P < 0.05$.

Results

After 35 days of trecadrine administration to the basal group, galactose uptake by the intestinal tissue decreased significantly (by 19%, $P < 0.05$), while no changes were observed in sucrase and maltase activities (Table 1).

The inhibitory effect of trecadrine on galactose absorption was also found in in-vitro conditions. Thus, data show that galactose uptake by intestinal rings (5.20 ± 0.14 nmol (mg wet weight)$^{-1}$, control conditions, $n = 21$) was significantly decreased ($P < 0.001$) by the presence of 10^{-5} M trecadrine in the incubation medium (4.53 ± 0.13 nmol (mg wet weight)$^{-1}$, $n = 22$).

A diabetic state was reached in alloxan-treated rats, since plasma glucose levels at the end of the experimental trial were significantly increased (from 5.05 ± 0.50 to 17.59 ± 2.99 mmol L^{-1}, $P < 0.001$) in diabetic rats compared with controls. Moreover, trecadrine administration for three days normalised glycaemia in practical terms (it decreased plasma glucose from 17.59 ± 2.99 to 6.05 ± 1.22 mmol L^{-1}, $P < 0.001$) in diabetic treated rodents as compared with non-treated diabetic animals. In alloxan-treated rats, a clear enhancement of galactose intestinal absorption and of disaccharidase activities was obtained (Table 2). After trecadrine administration, the increasing effect on galactose absorption was significantly attenuated. Likewise, the treatment induced a significant improvement in the sucrase but not in the maltase activity.

Also, an obese state was reached after the animals were fed for 40 days on cafeteria diet. Thus, at the end of the trial, both body weight and body-fat composition were significantly increased (from 263 ± 10 to 312 ± 11 g body weight, $P < 0.01$; and from 7.0 ± 1.0 to 16.3 ± 0.5% fat, $P < 0.001$; respectively) in obese rats as compared with controls. Trecadrine administration for 35 days significantly decreased these values (from 312 ± 11 to 290 ± 10 g, $P < 0.05$; and from 16.3 ± 0.5 to 6.6 ± 0.5% fat, $P < 0.001$, respectively).

The hypercholesterolaemic diet increased total cholesterol (from 2.55 ± 0.28 to 10.76 ± 1.43 mmol L^{-1}, $P < 0.01$) and induced a reduction in HDL-cholesterol levels (from 2.61 ± 0.23 to 1.40 ± 0.32 mmol L^{-1}, $P < 0.05$) as compared with controls. Treatment with trecadrine produced no beneficial effects in lipid profile (total cholesterol: 8.13 ± 1.72 mmol L^{-1}; HDL: 1.26 ± 0.14 mmol L^{-1}).

In both diet-induced obese and hypercholesterolaemic models, galactose uptake by the intestinal rings and disaccharidase activities (Table 2) was significantly decreased. Trecadrine oral administration had no effect on galactose absorption and significantly diminished the inhibitory effect on sucrase activity (Table 2).

Discussion

Recent studies have shown that β_3-adrenergic agonists can be useful for the therapy of diabetes, hypercholesterolaemia and obesity, due to their hypoglycaemic and lipolytic properties (Howe 1993). Also, it is known that those disorders cause alterations in the absorptive and digestive processes, and that there is involvement of α- and β-adrenoceptors in nutrient absorption (McIntyre et al 1993). This influence is commonly explained by a direct effect on gastrointestinal motility acting via smooth muscle tone, where the receptors are mainly located (β-agonists) or by inhibiting neurotransmitter release (α-agonists). There is also evidence for an α-mediated enhancement of net absorption and a reduction in net secretion (McIntyre et al 1992). Finally, in recent studies (reviewed by Manara et al 1995), β_3-adrenergic agonists have been reported to be potent inhibitors of intestinal motility, causing relaxation in circular intestinal smooth muscle. Nevertheless, the idea of a β-adrenoceptor-mediated modulation of nutrient uptake by a direct effect on mucosal absorption can not be discarded, although there are no firm data supporting this hypothesis.

Experimentally-induced diabetes has been associated with gastrointestinal mucosal hypertrophy and a subsequent enhancement of nutrient transport. This increase in intestinal hexose uptake has been attributed to an enhancement in glucose carrier maximal transport capacity, as a consequence of the recruitment of sodium-glucose co-transporters into previous non-transporting intervillus regions, independently of the hypertrophy of specific mucosal regions. In this way, glucose transporters (GLUT2 and GLUT5) and sodium-glucose co-transporter (SGLT1) content and mRNA are increased in enterocytes isolated from diabetic animals in both jejunum and ileum (Fedorak et al 1991).

Sucrase activity shows a rise along the villus-crypt axis during the early phase of induction of alloxan diabetes in rats

Table 1. Intestinal absorption (galactose uptake) and digestion functions (sucrase and maltase activities) as affected by oral trecadrine administration for 35 days in female Wistar rats.

	Control	Trecadrine (1 mg kg^{-1} day^{-1})
Galactose uptake (μmol (g wet weight)$^{-1}$)	5.3 ± 0.3	4.3 ± 0.2*
Sucrase activity (μmol hydrolysed (mg protein)$^{-1}$ min^{-1})	533 ± 20	588 ± 28
Maltase activity (μmol hydrolysed (mg protein)$^{-1}$ min^{-1})	1491 ± 25	1493 ± 37

Each value represents the mean ± s.e. *$P < 0.05$ when compared with control group.

Table 2. Effect of diabetes, obesity and hypercholesterolaemia on jejunal galactose absorption and on sucrase and maltase activities and the influence of trecadrine, in rats.

Physiological condition	Galactose absorption (μmol (g wet weight)$^{-1}$)		Maltase activity (μmol hydrolysed (mg protein)$^{-1}$)		Sucrase activity (μmol hydrolysed (mg protein)$^{-1}$ min^{-1})	
	Untreated	Trecadrine	Untreated	Trecadrine	Untreated	Trecadrine
Diabetes	74·3***	35·1**$^{+}$	28·4**	24*	82·7**	62**$^{+}$
Obesity	−22·5*	−22*	−17*	−15·1*	−51·4*	−15*$^{+}$
Hypercholesterolaemia	−17**	−10	−32*	−16	−11*	15*$^{+}$

Values in control rats were taken as 0. *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$, compared with controls. $^{+}P < 0.05$ compared with untreated group.

(Nakabou et al 1985). Parallel to this increase, it has been observed that there is an increment in sucrase-isomaltase mRNA abundance, apparently mediated by the stabilisation of mRNA (Hoffman & Chang 1992), or as a consequence of a decrease in the enzyme degradation (Olsen & Korsmo 1977). In this way, Levine et al (1982) proposed that hyperglycaemia could increase glucose diffusion from the blood, through the paracellular way, into the intestinal lumen, with the result of a stimulation of sucrase activity.

The results of our studies in diabetic rats are in agreement to previous data, since an increase in sugar digestion and absorption was found. The inhibition of this rise after Trecadrine administration could be explained by the normalisation of glycaemia. In this way, insulin treatment has been proved to revert the increased glucose transporter expression in diabetic animals, being the effect also explained by a state of normoglycaemia (Burant et al 1994). Moreover, there are no data supporting a direct action of insulin in glucose transporter's expression and activity, and, as Trecadrine action apparently does not have a direct relationship with changes in insulinaemia (Barrionuevo et al 1996), it can be suggested that insulin may not be as important as glycaemia in intestinal glucose uptake regulation. Furthermore, the partial normalisation observed in disaccharidase activities after Trecadrine administration could be mainly attributed to the reduction in the diabetic status.

It has been extensively reported that a high-carbohydrate diet increases hexose absorption across the enterocyte membranes (Ferraris et al 1993), which has been explained by an increase in the number of glucose carriers in both brush-border and basolateral membranes (Miyamoto et al 1993). Moreover, it has been reported that brush-border membrane vesicles prepared from butter-fat-fed animals (rich in saturated fatty acid) show a decrease in Na^{+}-dependent D-glucose transport (Brasitus et al 1989). In our trial, both the cafeteria and the hypercholesterolaemic diets can be considered high-fat, low-carbohydrate as compared to the animals fed on the control diets. Our results indicate a reduction in jejunal galactose absorption in both cases, which could be related both to the lower carbohydrate content in the diet and to the higher percentage of saturated fatty acids as compared with their respective controls.

Previous results have confirmed that high-fat diets severely reduce sucrase and isomaltase activities in brush-border rat jejunum (Thomsen et al 1983), probably mediated by an increase in the pancreatic proteinase-induced degradation of these microvillar disaccharidases (Goda & Takase 1994). It has been demonstrated that sucrase-isomaltase mRNA levels are enhanced in rats fed a diet rich in carbohydrates (Broyart et al 1990), which involves an increase in sucrase activity. In the present experimental trial, as the hypercholesterolaemic and obese groups received a lower carbohydrate/fat ratio as compared to their control groups, our results show a subsequent decrease in sucrase activity in both the two groups, in agreement with other authors (Goda & Takase 1994).

Trecadrine treatment in both diet-induced obese and hypercholesterolaemic groups produces only a partial normalisation in disaccharidase activities, which is more evident on sucrase activity. This could be explained by the fact that Trecadrine, in common with other β_3-adrenergic agonists, elicits a potent lipomobilising effect (Milagro et al 1996) with subsequent changes in serum lipid composition, which could affect membrane lipid distribution. In this context, variations in biophysical characteristics of the enterocyte membranes have been reported to modify some intestinal enzyme activities (Stenson et al 1989).

Finally, Trecadrine administration does not significantly attenuate the inhibition of the galactose uptake by intestinal rings in hypercholesterolaemic and obese diet-induced rats, which could be explained by a direct inhibitory effect of the drug on sugar intestinal absorption. Actually, Trecadrine administration to a basal group caused a significant decrease in jejunal galactose uptake. This effect on sugar absorption has been found also in in-vitro experiments when Trecadrine was added to the incubation medium. These results could not be explained if the β_3-adrenergic agonist action on gastrointestinal tract only occurs on the inhibition of the motor or the vascular function, which points to a direct effect of β_3-adrenergic agonists on carbohydrate intestinal absorption.

References

Barrionuevo, M., Milagro, F. I., Cenarruzabeitia, E., Martínez, J. A. (1996) Potential anti-diabetic applications of a new molecule with affinity for β_3-adrenoceptors. Life Sci. 59: 141–146

Barry, M. K., Aloisi, J. D., Yeo, C. J. (1993) Luminal adrenergic agonists modulate ileal transport by a local mechanism. J. Surg. Res. 54: 603–609

Bradford, M. M. (1976) A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal. Biochem. 72: 248–254

Brasitus, T. A., Dudeja, P. K., Bolt, M. J., Sitrin, M. D., Baum, C. (1989) Dietary triacylglycerol modulates sodium-dependent D-glucose transport, fluidity and fatty acid composition of rat small

intestinal brush-border membrane. Biochim. Biophys. Acta 979: 177–186

Broyart, J.P., Hugot, J.P., Perret, C., Porteu, A. (1990) Molecular cloning and characterization of a rat intestinal sucrase-isomaltase cDNA: regulation of sucrase-isomaltase gene expression by sucrose feeding. Biochim. Biophys. Acta 1087: 61–67

Burant, C. F., Flink, S., DePaoli, A. M., Chen, J., Wen-Sen L., Hedigger, M., Buse, J. B., Chang, E. B. (1994) Small intestine hexose transport in experimental diabetes. J. Clin. Invest. 93: 578–585

Dahlqvist, A. (1964) Method for assay of intestinal disaccharidases. Anal. Biochem. 7: 18–25

Evans, B. A., Papaioannou, M., Bonazzi, V. R., Summers, R. J. (1996) Expression of β_3-adrenoceptor mRNA in rat tissues. Br. J. Pharmacol. 117: 210–216

Fedorak, R. N., Cheeseman, C. I., Thomson, B. R., Porter, V. M. (1991) Altered glucose carrier expression: mechanism of intestinal adaptation during streptozocin-induced diabetes in rats. Biochem. Biophys. Res. Commun. 181: 1110–1117

Ferraris, R. P., Vinnakota, R. R. (1995) Intestinal nutrient transport in genetically obese mice. Am. J. Clin. Nutr. 62: 540–546

Ferraris, R. P., Casirola, D. M., Vinnakota, R.R. (1993) Dietary carbohydrate enhances intestinal sugar transport in diabetic mice. Diabetes 42: 1579–1587

Goda, T., Takase, S. (1994) Dietary carbohydrate and fat independently modulate disaccharidase activities in rat jejunum. J. Nutr. 124: 2233–2239

Hoffman, L. R., Chang, E. B. (1992) Altered regulation of regional sucrase-isomaltase expression in diabetic rat intestine. Am. J. Physiol. 262: G983–G989

Howe, R. (1993) β_3-Adrenergic agonist. Drugs Fut. 18: 529–549

Idoate, I., Mendizábal, M. V., Urdaneta, E., Larralde, J. (1996) Interactions of cephadrine and cefaclor with the intestinal absorption of D-galactose. J. Pharm. Pharmacol. 48: 645–650

Levine, G. M., Shiau, Y. F., Deren, J. A. (1982) Characteristics of intestinal glucose secretion in normal an diabetic rats. Am. J. Physiol. 242: G455–G459

Lugea, A., Barber, A. and Ponz, F. (1994) Inhibition of D-galactose and phenylalanine transport by $HgCl_2$ in rat intestine in-vitro. Rev. Esp. Fisiol. 50: 167–174

Manara, M., Croci, T., Landi, M. (1995) β_3-Adrenoceptors and intestinal motility. Fundam. Clin. Pharmacol. 9: 332–342

McIntyre, A. S., Thompson, D. G., Burnham, W. R., Walker, E. (1992) The effect of α_1-adrenoreceptor agonist and antagonist administration on human upper gastrointestinal transit and motility. Aliment. Pharmacol. Ther. 6: 415–426

McIntyre, A. S., Thompson, D. G., Burnham, W. R., Walker, E. (1993) The effect of β-adrenoreceptor agonists and antagonists on fructose absorption in man. Aliment. Pharmacol. Ther. 7: 267–274

Milagro, F. I., Gómez-Ambrosi, J., Martínez, J. A. (1996) Lipomobilizating and hypoglycaemic effects of a β_3-adrenergic agonist in diabetic rats. Int. J. Obesity 20: 87

Miyamoto, K., Hase, I., Takagi, T., Fuji, T., Taketani, Y., Minami, H., Oka, T., Nakabou, Y. (1993) Differential responses of intestinal glucose transporter mRNA transcripts to levels of dietary sugars. Biochem. Biophys. Res. Commun. 183: 626–631

Nakabou, Y., Ikeuchi, K., Minami, H., Hagihira, H. (1985) Changes in brush-border enzyme activities of intestinal epithelial cells isolated from the villus-crypt axis during the early phase of alloxan diabetes in rats. Experientia 41: 482–484

Nashiro, K., Murakami, K., Mimura, G. (1992) Diurnal variation and increase of disaccharidase activity in diabetic rats. J. Nutr. Sci. Vitaminol. Tokyo 38: 265–276

Olsen, W. A., Korsmo, H. (1977) The intestinal brush border membrane in diabetes. Studies of sucrase-isomaltase metabolism in rats with streptozotocin diabetes. J. Clin. Invest. 60: 181–188

Planas, B., Pons, S., Nicolau, M. C., López-García, J. A., Rial, R. (1992) Morphofunctional changes in gastrointestinal tract of rats due to cafeteria diet. Rev. Esp. Fisiol. 48: 37–43

Shirazi-Beechey, S. P., Davies, A. G., Tebbutt, K., Dyer, J., Ellis, A., Taylor, C. J., Fairclough, P., Beechey, R.B. (1990) Preparation and properties of brush-border membrane vesicles from human small intestine. Gastroenterology 98: 676–685

Stenson, W. F., Seetharam, B., Talkad, V., Pickett, W., Dudeja, P., Brasitus, T. A. (1989) Effects of dietary fish oil supplementation on membrane fluidity and enzyme activity in rat small intestine. Biochem. J. 263: 41–45

Thollander, M., Svensson, T. H., Hellstrom, P. M. (1996) β-Adrenoceptors regulate myoelectric activity in the small intestine of rats: stimulation by β_2 and inhibition by β_3 subtypes. Neurogastroenterol. Mot. 8: 143–151

Thomsen, L. T., Tasman-Jones, C., Maher, C. (1983) Effects of dietary fat and gel-forming substances on rat jejunal disaccharidase levels. Digestion 26: 124–130

Zulet, M. A., Martínez, J. A. (1995) Corrective role of chickpea intake on a dietary-induced model of hypercholesterolaemia. Plant Foods Hum. Nutr. 48: 269–277