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Tissue localization of threonine oxidation in pigs

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Two experiments were designed to determine the tissue distribution of threonine oxidation through the threonine dehydrogenase (*EC* 1.1.1.103) pathway in pigs. The first experiment was conducted on eleven Piétrain × Large White piglets. The piglets were slaughtered at 5, 12 or 20 kg after 1 h of infusion with L-[U-^{14}C]threonine (55 kBq/kg) mixed with unlabelled threonine (100 mg/kg). In the second experiment, four Piétrain × Large White and four Large White piglets (10 kg body weight) were infused with L-[1-^{13}C]threonine (50 mg/kg) mixed with 50 mg/kg unlabelled threonine for 1 h, then killed for tissue sampling. In the two experiments, threonine dehydrogenase specific activity and threonine and glycine specific radioactivities and enrichments were measured in several tissues and in plasma. The higher level of labelling of threonine in the pancreas than in the liver suggested either a lower protein degradation rate or a faster rate of threonine transport in the liver than in the pancreas. Threonine dehydrogenase activity was found only in the liver and the pancreas. Whereas liver and pancreas threonine dehydrogenase specific activities were similar, glycine specific radioactivity and enrichment were 12- to 14-fold higher in the pancreas than in the liver. This is probably the consequence of a higher production rate of glycine from sources other than threonine (protein degradation, *de novo* synthesis from serine) in the liver than in the pancreas. Our results showed that Large White pigs could oxidize more threonine than Piétrain × Large White pigs. This could be related to the difference in growth performance and dietary N efficiency for protein deposition between these two genotypes.

Threonine: Amino acids: Genotype: Pigs

Threonine is often a limiting amino acid for growth and maintenance in diets consumed by pigs. Consequently, conservation of threonine from oxidation is crucial to maintain protein synthesis and deposition. In pigs, threonine dehydrogenase (*EC* 1.1.1.103; TDG) is the main enzyme involved in threonine degradation (Ballèvre *et al.* 1990; Le Floc'h *et al.* 1995). This enzyme catalyses the oxidation of L-threonine to 2-amino-3-oxobutyric acid which is spontaneously decarboxylated to give aminoacetone (Green & Eliott, 1964) or glycine and acetyl CoA when TDG is coupled with 2-amino-3-oxobutyrate CoA ligase (*EC* 2.3.1.29) (Tressel *et al.* 1986). *In vivo* the production of glycine is predominant (Bird *et al.* 1984), but the measurement of aminoacetone production is used for *in vitro* TDG specific activity determination (Hartshorne & Greenberg, 1964). However, *in vitro* enzyme activity does not always reflect *in vivo* oxidation since it does not integrate certain regulatory processes such as transport into the cell or inter-organ flux. It is possible to determine *in vivo* threonine oxidation through the infusion of labelled threonine and measurement of labelled CO_2 (Kang-Lee & Harper, 1978) but the measurement of glycine (Ballèvre *et al.* 1990) production rate is the preferred approach.

The main problems with this latter technique are the compartmentation of amino acid metabolism and the difficulty in determining the true amino acid precursor pool for oxidation. When the plasma was chosen as the precursor pool, this led to an erroneous

estimation of the absolute value of oxidation since, in pigs, threonine oxidation occurs mainly in the liver (Ballèvre *et al.* 1990). However, we have shown in a previous experiment that threonine oxidation to glycine also takes place in other tissues (Le Floc'h *et al.* 1995) and that the contribution of the liver to total threonine oxidation decreases with the level of dietary threonine as a consequence of a reduced threonine transport into the liver (Le Floc'h *et al.* 1996). It is necessary to determine which tissues are involved in this degradative pathway in order to improve the model used to estimate the true rate of *in vivo* threonine oxidation to glycine and to determine the partition of oxidation between the liver and the extra-hepatic tissue(s) according to the level of dietary threonine supply.

In the present paper we report the results of two experiments in which we have measured and compared, in different tissues, the TDG activity and glycine enrichment or specific radioactivity after the infusion of labelled threonine. Moreover, we have compared threonine oxidation levels in two genotypes of pigs which are known to present different growth performances and responses to nutrient supply.

MATERIALS AND METHODS

Chemicals

L-[U-^{14}C]Threonine (6·7 GBq/mmol) was provided by ICN France (Orsay, France) and L-[1-^{13}C]threonine by Isotec France (Saint Quentin, France). All other chemicals, except aminoacetone (synthesized in our laboratory), were purchased from Sigma (St Louis, MO, USA) or Boehringer (Mannheim, Germany).

Animals and infusion protocol

Expt 1. Twelve female Piétrain × Large White pigs from the Institut National de la Recherche Agronomique (INRA) herd were used in this experiment. These pigs were selected in order to weigh approximately 5 (4·39, SE 0.11), 12 (11·77, SE 0·18) or 20 (20·44, SE 0·65) kg at the time of the infusions. A catheter was inserted under anaesthesia through each pig's jugular vein into the cranial vena cava as previously described (Le Floc'h *et al.* 1995). The animals were fed twice daily (Table 1) and, in order to ensure a correct and constant nutritional status (Cortamira *et al.* 1991), piglets weighing 5 kg were fed on a liquid diet through a gastric tube. The dry diet was mixed with water (1 : 3, w/w) and was delivered in liquid form with a syringe. A solution of threonine (100 mg/kg body weight (BW)) combined with L-[U-^{14}C]threonine (55 kBq/kg BW) was prepared in sterile saline (9 g NaCl/l) and passed through a 0·22 μm filter before use. At 2 d after surgical preparation, this solution was infused via the jugular catheter for 1 h from 30 min after the meal. Blood (20 ml) was sampled from the catheter and the piglets were killed with a lethal dose of pentobarbital. Liver, pancreas, kidney, spleen, lung, duodenum, heart and muscle were sampled and frozen in liquid N_2 and stored at $-80°$.

Expt 2. Four pairs of female littermate Piétrain × Large White and Large White pigs were selected at an average weight of 10 kg. They were surgically prepared as described earlier. The infusion protocol was similar to the protocol of the first experiment (infusion of 100 mg L-threonine/kg BW) except that L-[U-^{14}C]threonine was replaced by L[1-^{13}C]threonine (50 mg/kg BW). Only one pig of each pair was infused and the other one was slaughtered at the same time in order to determine threonine and glycine basal ^{13}C abundance in the different tissues.

Table 1. *Composition of the diets (g/kg)*

Ingredients	Weaner phase 1*	Weaner phase 2*
Barley	414	145
Wheat	–	233
Maize	–	280
Soyabean meal, 50	200	–
Soyabean meal, 48	–	255
Sweet dehydrated whey	200	–
Skimmed milk powder	80	–
Fish meal	50	50
Tallow	20	–
Dicalcium phosphate	18	18
Limestone	11	10
Iodized salt	–	4
L-Threonine	0·8	–
DL-Methionine	1·2	–
Trace mineral and vitamin premix†	5	5

* Weaner phase 1 was offered to 5 kg piglets and weaner phase 2 to 12 and 20 kg piglets.
† For details of premix see Le Floc'h *et al.* (1996).

In the two experiments, the amount of infused threonine corresponded to approximately one quarter of ingested threonine per day. Unlabelled threonine was included in the infusion of labelled threonine in order to (1) reduce the difference in the precursor pool (threonine) labelling in different tissues and (2) increase glycine production from threonine and, consequently, to improve the detection of TDG in tissues where threonine oxidation is low.

Analytical procedure

Tissue samples (10 g) and plasma (20 ml) were homogenized in 40 ml TCA (100 g/l) and centrifuged for 20 min at 133 ***g***. Amino acids from the supernatant fraction were then purified through cation exchange resin (AG 40W-X8, 100-200 mesh, Biorad, Ivry Sur Seine, France) and eluted with 4 M-NH_4OH. NH_3 was evaporated under vacuum and amino acids were recovered in a small volume (1 ml) of sodium acetate buffer pH 2·2, for liquid chromatography, or in water, for mass spectrometry analysis.

Approximately half the tissue amino acids sample and 75 % of plasma amino acids were saved for glycine and threonine specific radioactivities measurement. Glycine and threonine were purified by liquid chromatography (Multichrom amino acid analyser, Beckman, München, Germany). Separation was performed on cation exchange resin (CK 10S, Mitsubichi; Prolabo, Gradignan, France) with a 10 × 400 mm column. Amino acids were eluted (flow rate 57 ml/h, 57°) with sodium citrate buffer (pH 2·92, 70 g/l and 120 g/l isopropanol for glycine and threonine respectively) and collected with an automatic fraction collector (Gilson 202, Villiers Le Bel, France). Under these conditions the retention times are approximately 75 min and 130 min for threonine and glycine respectively. There was no detectable radioactivity in serine, alanine or asparagine which could have contaminated the threonine and glycine fractions. Glycine and threonine concentrations in the eluate were determined by liquid chromatography and ninhydrin colouration. Radioactivity of the pooled fractions was measured by liquid scintillation counting (Ultima gold or Hionic Fluor, Packard, Downers Grove, IL, USA) on a liquid scintillation analyser (Tri-Carb 1300 TR, Packard).

Threonine enrichment was determined by GC–mass spectrometry (GC–MS) in electron impact ionization mode (GC 8060 chromatograph coupled to a VG Platform II, Fisons Instruments, Altrincham, Ches.) after derivatization of the sample with N-methyl-N-*tert*-butyldimethylsilyl trifluoroacetamide (MTBSTFA). Amino acid derivatives were separated on a 30 m, 0·25 mm inner diameter (i.d.) and 0·1 μm film thickness capillary column (DB 5-MS, J&W Scientific, Courtaboeuf, France). He gas was used as a carrier (flow 1·2 ml/min). Best chromatographic resolution of the amino acid derivatives was observed using a temperature programme which started at 100° (2 min), rose at the rate of 10°/min to 250° and was kept constant for 6 min. Selected ion monitoring mode (SIM) was used to scan
the specific ions masses (M-57) and (M-57) + 1 for tri-*tert*-butyl dimethylsilyl threonine (m/z = 404 and 405 respectively). The peak areas ratios (405 : 404) were calculated for each tissue in the infused pig ($R_{enriched}$) and the control pig (R_0). Threonine enrichment was calculated with the conventional formula:

$$\text{enrichment (APE, \%)} = 100 \times (R_{enriched} - R_0)/[1 + (R_{enriched} - R_0)].$$

Due to its lower enrichment, ^{13}C content of glycine was determined by GC–combustion–isotope ratio mass spectrometry (GC–C–IRMS) (GC 8130 chromotograph interfaced with a VG Isochrom mass spectrometer, Fisons Instruments) after derivatization with ethyl chloroformate according to the method described by Husek (1991). Separation of amino acid derivatives was performed on a DB 1701 capillary column (J&W Scientific) with the following characteristics: 30 m, 0·25 mm i.d. and 0·25 μm film thickness. In this study, the best chromatographic conditions were as follows: injector temperature was 240°, the column was held isothermal at 120° for 4 min after injection, then the temperature was increased to 190° at the rate of 20°/min, maintained constant for 4 min, and further elevated at the rate of 30°/min to 250° which was held for 4 min.

TDG specific activity was measured as the rate of aminoacetone formation *in vitro*, as previously described (Le Floc'h *et al.* 1994).

Statistical analysis

Data were submitted to ANOVA according to the general linear models (GLM) procedure of the Statistical Analysis System (1989). The effects of weight and genotype were tested against the residual variation between pigs. Tissues and plasma values were compared against the residual variation within animal. When a statistical difference was established by Fisher–Snedecor ($P < 0·05$), means were compared by Student's t test and declared different at $P < 0·05$.

RESULTS

Threonine dehydrogenase specific activity

TDG activity was found in pancreas and in liver but not in the other tissues. In pigs infused with L-[U-^{14}C]threonine (Fig. 1(a); Expt 1), liver TDG specific activity was not affected by the weight of pigs whereas pancreas TDG activity was higher in 20 kg pigs than in 12 kg pigs. Enzyme specific activity was significantly higher in the liver than in the pancreas in 10 kg pigs ($P = 0·02$) whereas the reverse was true in 20 kg pigs ($P < 0·01$). Results of the second experiment (Fig 1(b)) showed that liver TDG specific activity was similar in Large White and Piétrain × Large White pigs whereas pancrease TDG activity was significantly higher in Large White than in Piétrain × Large White pigs ($P < 0·01$).

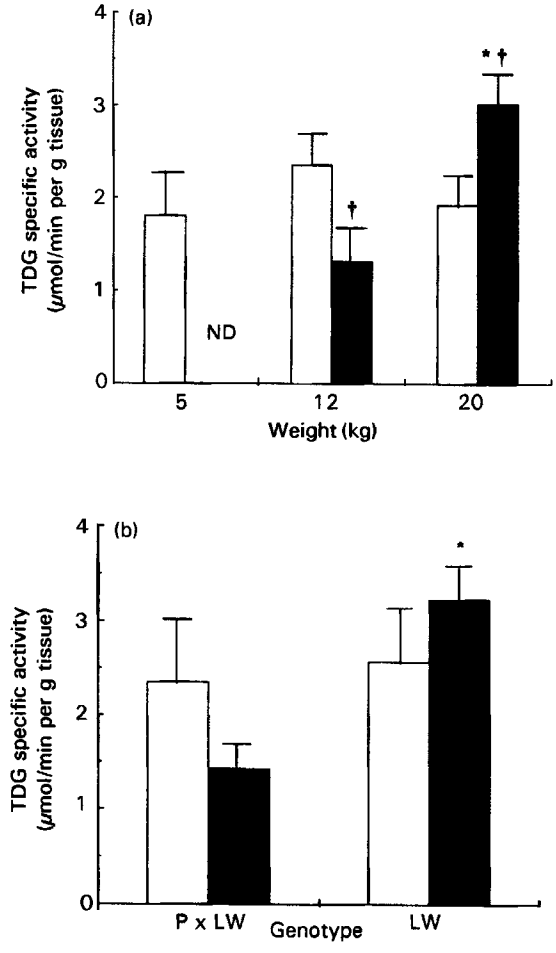

Fig. 1. Liver (□) and pancreas (■) L-threonine dehydrogenase (TDG) specific activity, expressed as the rate of aminoacetone formation, in pigs infused with (a) L-[U-^{14}C]threonine or (b) L-[1-^{13}C]threonine. Values are means for each group with their standard errors represented by vertical bars. Panel (a): *n* 4 for 5 and 20 kg live weight pigs and *n* 3 for 12 kg live weight pigs; panel (b): *n* 4. Panel (a): * mean values were significantly different between stages of growth, $P < 0.05$; † mean value was significantly different from that for the liver, $P < 0.05$. Panel (b): * mean value was significantly different from that for P × LW pigs, $P < 0.05$. ND, not determined; LW, Large White; P, Piétrain.

Threonine enrichment and specific radioactivity

The values of threonine specific radioactivity and enrichment are presented in Table 2. The results obtained with both tracers were similar. Because there was no effect of age or of genotype on threonine enrichment and specific radioactivity, the results are presented as means for each tissue. Threonine specific radioactivity was significantly higher in the pancreas than in the other tissues ($P < 0.02$). In pigs infused with L-[1-^{13}C]threonine, threonine enrichment was significantly higher in the pancreas than in the liver and the heart ($P < 0.05$) and was close to plasma, kidney and muscle enrichments. Threonine enrichment measured in brain was much lower than in the other tissues ($P = 0.0001$).

Table 2. *Threonine enrichments (E) and specific radioactivities (SR) in tissues and plasma of pigs after the infusion of* L-*[1-*^{13}*C]threonine or* L-*[U-*^{14}*C]threonine*†

(Mean values with their standard errors for eight (enrichment) or twelve (specific radioactivity) piglets)

Infusion...	L-[1-^{13}C]Threonine E (APE)		L-[U-^{14}C]Threonine SR (Bq/μmol)	
Tissue	Mean	SE	Mean	SE
Liver	20·1*	1·0	33·9*	1·1
Pancreas	22·2	0·7	40·9	1·2
Lung	20·4	0·8	33·9*	1·0
Kidney	21·8	0·7	35·5*	0·9
Spleen	20·6	0·6	35·5*	0·8
Heart	19·4*	0·75	31·1*	1·5
Brain	10·5*	0·9	ND	
Duodenum	21·1	0·9	35·5*	1·7
Muscle	22·1	0·8	35·8*	1·5
Plasma	22·5	0·6	41·2	1·0

APE, atom percent excess; ND, not determined.

* Mean values were significantly different from plasma value ($P < 0·05$).

† For details of procedures, see pp. 594–596.

Glycine enrichment and specific radioactivity

In all tissues, except in the liver and in the pancreas, glycine labelling was not different from plasma values. Glycine enrichment and specific radioactivity were significantly higher in the liver and in the pancreas than in the other tissues and in the plasma ($P = 0·0001$). Glycine specific radioactivity and enrichment were 12- to 14-fold higher in the pancreas than in the liver (Figs. 2(a) and 2(b)). Pancreatic and liver glycine specific radioactivities were significantly affected by the weight of pigs. In Expt 2, glycine enrichment in the pancreas was significantly lower ($P = 0·001$) in Piétrain × Large White than in Large White pigs but the same difference was not significant in the liver or in the plasma ($P = 0·7$). However, if the pancreas was excluded from the statistical analysis due to the higher variance, the difference between breed appeared to be significant in the liver and the plasma ($P < 0·05$).

DISCUSSION

The aim of the present study was to determine the tissue localization of threonine oxidation in the pig using simultaneously an *in vitro* (measurement of TDG specific activity) and an *in vivo* (infusion of labelled threonine) approach. The second approach was not quantitative but was presumed to be more sensitive for the detection of TDG activity. We assumed that a tissue could oxidize threonine if TDG activity was detected *in vitro* and if glycine enrichment or specific radioactivity was higher in this tissue than in the plasma during labelled threonine infusion. Our purpose was to reduce differences in intracellular oxidation precursor pool (threonine) labelling in order to limit variations of glycine labelling associated with the experimental procedure. The infusion technique was intermediate between the flooding-dose and the continuous-infusion techniques and it is important to point out that the results obtained with this technique were only qualitative because threonine oxidation rate based on steady-state equations could not be used in this

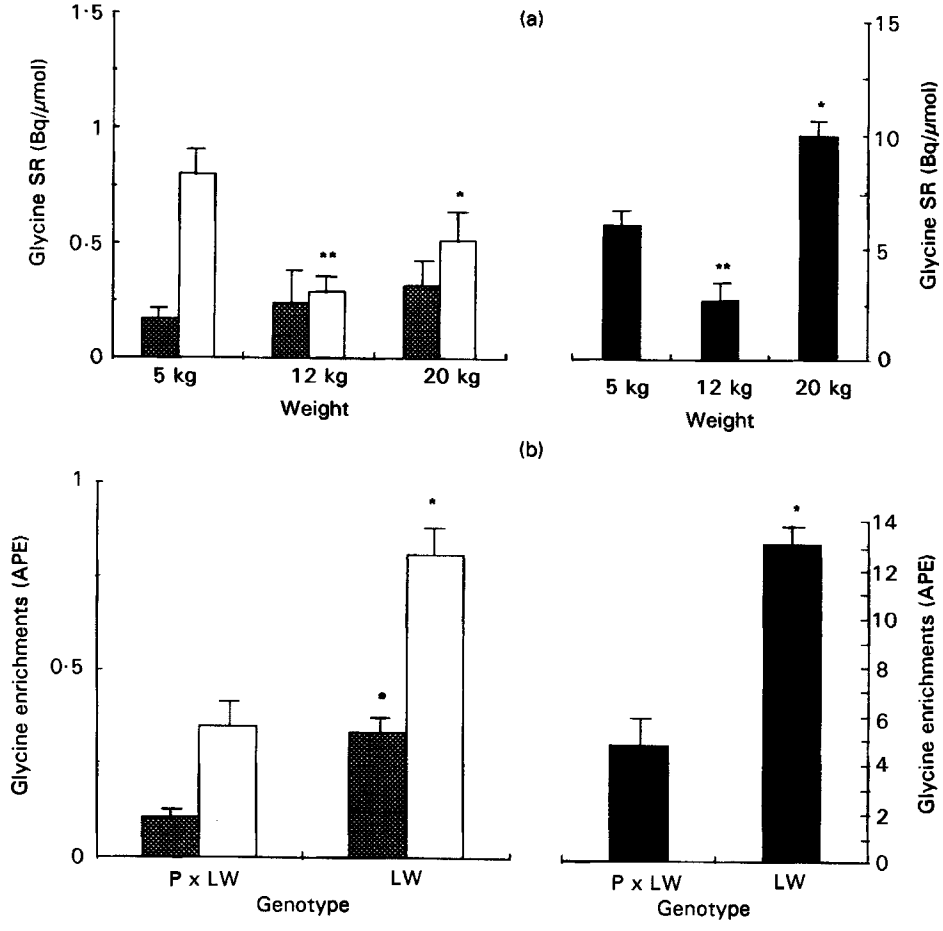

Fig. 2. Glycine (a) specific radioactivity (SR) and (b) enrichment measured in the plasma (▩), the liver (□) and the pancreas (■) of pigs infused with (a) L-[U-^{14}C]threonine or (b) L-[1-^{13}C]threonine. Values are means for each group, with their standard errors represented by vertical bars. Panel (a): n 4 for 5 and 20 kg live weight pigs and n 3 for 12 kg live weight pigs; panel (b): n 4. Panel (a): mean values were significantly different from those for 5 kg pigs: * $P < 0{\cdot}05$, ** $P < 0{\cdot}01$. Panel (b): * mean values were significantly different from those for P × LW pigs, $P < 0{\cdot}05$. APE, atom percent excess; LW, Large White; P, Piétrain.

experiment. Continuous infusion for 1 h was preferred to a bolus injection of threonine in order to avoid a large loss of tracer in the urine. Simultaneous infusion of labelled and unlabelled threonine, partly flooding the free pools, helped to minimize the consequence of protein degradation on intracellular threonine labelling and, since pigs were in an absorptive state as in our previous experiments (Le Floc'h *et al.* 1995, 1996), the effect of the dilution with dietary threonine in the liver and in the intestine. An associated advantage of unlabelled threonine infusion was to increase glycine production in tissues where the contribution of threonine oxidation to glycine flux is low. Since TDG activity was not induced by an excess of threonine (Le Floc'h *et al.* 1996), the infusion of unlabelled threonine probably had no effect on qualitative estimation and comparison of oxidative potential between pigs.

Compartmentation of threonine metabolism

Intracellular amino acid enrichment depends both on the rate of exchange between plasma and tissue and on the dilution of labelled amino acids by unlabelled amino acids released by protein degradation. As discussed earlier, the infusion of unlabelled threonine should have reduce the difference in threonine labelling between tissues. Indeed, the results presented in Table 2 show that threonine enrichments and specific radioactivities were similar in the different tissues with two exceptions: the pancreas and the brain.

After 1 h of L-[U-^{14}C] or [1-^{13}C]threonine infusion the enrichment and specific radioactivity of free threonine in the pancreas were significantly higher than in the liver and close to the plasma value as previously noticed after 10 h of infusion (Le Floc'h *et al.* 1995). This result was unexpected since the turnover rate of pancreatic proteins is very fast compared with that of muscle (Simon *et al.* 1982; Ponter *et al.* 1994) and might have been expected to induce a lower pancreatic threonine enrichment. However, even after 6 h of L-[1-^{14}C]lysine or L-[1-^{14}C]leucine infusion, free lysine and leucine specific radioactivities measured in the pancreas of fasted pigs were higher than those of liver and close to the values measured in the other tissues (Simon *et al.* 1978, 1982). In this last case the low values of liver free lysine and leucine specific radioactivities were interpreted as the consequence of an increase in liver protein breakdown during fasting (Garlick *et al.* 1973), but this could not be so in our experiment where pigs were in an absorptive state. It was equally improbable that the high value of pancreatic threonine enrichment observed in the present experiment was due to tracer recycling as the infusion was for only 1 h.

According to Simon *et al.* (1983), and more recently to Leterme *et al.* (1993), free amino acid labelling in the pancreatic juice is close to that of plasma and blood during a continuous tracer infusion. In the rat infused with [^{3}H]leucine, Girard-Globa *et al.* (1980) showed that labelling of the precursor pool for pancreatic juice protein synthesis was close to that in plasma free amino acids. This is consistent with the fact that, during the infusion of labelled threonine, intra-tissue pancreas free threonine enrichment tended to equilibrate with plasma enrichment in the present as in previous experiments (Le Floc'h *et al.* 1995, 1996). Pancreatic juice protein synthesis is probably a major component of total protein synthesis activity in the pancreas. About 18 g protein is excreted daily by the pancreas of a 45 kg live weight pig (Corring, 1975). Consequently, amino acids entering the pancreas are little diluted since pancreatic juice protein degradation occurs in the intestine and recycling may occur in any tissues other than in the pancreas. Alternatively, threonine transport into the pancreas might be easier and faster than in the other tissues and particularly than in the liver. Conversely, the low value of threonine enrichment measured in the brain could be the consequence of the slow transport of threonine across the blood–brain barrier which prevents the negative effect of amino acid accumulation in the brain on food intake (Peng *et al.* 1973).

Tissue localization of threonine oxidation

Measurements of *in vitro* TDG specific activity in different tissues have shown that the enzyme is present in the liver and in the pancreas. No activity could be detected in the other tissues. This result was confirmed *in vivo* with the measurement of higher glycine enrichment and specific radioactivity in these tissues than in the plasma after the infusion of labelled threonine. It was in agreement with the data of previous experiments (Le Floc'h *et al.* 1995, 1996). The role of the pancreas in threonine metabolism has been described in the chicken by Davis & Austic (1982). In this species, TDG specific activity is fivefold

higher in the pancreas than in the liver. More recently, the same authors (Davis & Austic, 1994) showed that the enzyme was widespread in many tissues. In rats, TDG specific activities measured in the brain and in the kidney represented 10 and 30 % of liver activity when expressed per gram of tissue (Green & Eliott, 1964), but, taking into account the mass of the liver, 87 % of TDG activity appeared to be located in the liver. In our experiment, although *in vitro* TDG specific activities were similar in the liver and in the pancreas, total liver activity represented more than 90 % of total activity.

This result was not reflected either in values of glycine enrichment or in specific radioactivity in the pancreas which were twelve times higher than liver enrichment. It is clear that the high value of pancreas glycine enrichment could be only partly the consequence of the higher level of labelling of the threonine pool. In fact, the absolute value of *in vivo* threonine oxidation rate depends on glycine flux. On one hand, the value of liver glycine flux estimated by Ballèvre *et al.* (1991) was high and close to plasma glycine flux. On the other hand, in the pancreas, the part of glycine flux due to protein degradation was probably not as high as expected since, as discussed earlier, pancreatic juice protein is exported, degraded and recycled elsewhere. Moreover, in the pancreas, unlike in the liver, TDG might be the main pathway involved in glycine *de novo* synthesis. Therefore, the present results may remain consistent with the assumption that the liver was the tissue which oxidized the greatest amount of threonine *in vivo* when the supply of threonine is not limiting or important, as was the case during the infusion of 100 mg/kg BW.

The effect of weight and genotype on threonine oxidation

The comparison of TDG activity and glycine specific radioactivity measured in piglets of different weights (Expt 1, Fig. 1) showed that the changes in TDG specific activity and in glycine specific radioactivity were similar in the pancreas but not in the liver. Consequently, TDG activity was probably not limiting for liver threonine oxidation. This result was consistent with our hypothesis concerning the role of threonine transport as the limiting step for threonine oxidation in the liver but not in the pancreas (Le Floc'h *et al.* 1996). The lower values of liver and pancreas glycine enrichments measured in 12 kg pigs could be a consequence of an adaptation to undernutrition associated with weaning which occurred only 10 d before the infusion.

Our data showed an effect of genotype on pig oxidative capacities. Indeed, TDG specific activity and glycine enrichment measured in liver and pancreas were higher in Large White than in crossbred pigs. These genotypes differ in their growth performance and their ability to use dietary protein and amino acids efficiently for protein deposition. With the same energy and protein intake, crossbred pigs deposited 60 g/d more protein than Large White pigs (Quiniou, 1995). Previous results obtained from our laboratory have shown that muscle protein synthesis is 17 % faster in Large White pigs than in crossbred pigs (B. Sève and E. Capparo, unpublished results) probably reflecting higher protein turnover. Consequently, the higher oxidative potential in Large White pigs could be another aspect of a poor metabolic efficiency involving their inability to spare dietary amino acids for protein synthesis. On the other hand, this could signify that Large White pigs were able to clear excess threonine easily. The reason why the difference seemed to affect pancreatic TDG activity more dramatically is unknown. However, previous results have shown that the contribution of the pancreas to total threonine oxidation is more important when dietary threonine supply is low because of the reduced availability of threonine in the liver (Le Floc'h *et al.* 1996). Therefore, threonine basal oxidation rate,

representing part of the maintenance requirement, might be higher in Large White than in crossbred pigs.

In conclusion, the present data show that, in pigs, threonine oxidation takes place only in the liver and in the pancreas. The comparison of threonine enrichment in different tissues suggested that threonine was more rapidly taken up in the pancreas than in the liver. This result supports our previous hypothesis that threonine transport might be the limiting step controlling *in vivo* threonine oxidation in the liver, but not in the pancreas (Le Floc'h *et al.* 1996). On the other hand, we have shown that Large White pigs would be able to oxidize more threonine than crossbred pigs. This result is consistent with the difference in dietary protein and amino acid efficiency for protein deposition between these two genotypes.

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