To further analyse this greater inhibition of protein synthesis to DEX in old rats, protein synthesis was measured in isolated epitrochlearis incubated in the presence of ^{14}C phenylalanine. The effect of DEX in this case was a decrease in protein synthesis in old rats but not in adults, suggesting an intrinsic change during ageing in the effect of DEX on skeletal muscle.

Since DEX is a potent diabetogenic factor, in vitro protein synthesis was also measured in the presence of insulin (30 nM). DEX-treated rats showed an insulin-resistant state. This phenomenon was greater in old than in adult rats (–26 % DEX-treated versus control adult rats, –49 % DEX-treated versus control old rats, $P < 0.01$). Note that these results obtained in vitro in the presence of insulin are in total accordance with those obtained in vivo (*figure 1*).

If we consider all these findings, we can conclude that a higher responsiveness to glucocorticoids along with the establishment of a greater insulin-resistant state may explain muscle protein wasting during ageing.

Communication no. 31

The rapamycin-sensitive pathway is not involved in the impairment of skeletal muscle protein synthesis observed in diabetes-associated or glucocorticoid-induced insulin resistance. K. Grzelkowska, D. Dardevet, C. Sornet, M. Balage, J. Grizard (Unité d'étude du metabolisme azoté, Inra, Theix, 63122 Saint-Genès-Champanelle, France)

It has been shown that the PI-3 kinase/$p70^{S6K}$ signalling pathway was involved in insulin-regulated muscle protein synthesis (Dardevet et al., Endocrinology 137 (1996) 4087–4094). The aim of the present study was to examine the possible contribution of the $p70^{S6K}$ pathway in the impairment of skeletal muscle protein synthesis observed in the case of insulin resistance. We investigated the effect of rapamycin on insulin-stimulated protein synthesis in rat epitrochlearis muscle in vitro. Rapamycin is considered as a 'specific' inhibitor of transduction pathways including $p70^{S6K}$ (Brown et al., Nature 377 (1995) 441–446). The $p70^{S6K}$ protein level and $p70^{S6K}$ activity were analysed by western blotting and the immune complex kinase assay. Two models of insulin resistance were used: streptozotocin (STZ)-diabetic rats (3 days after intravenous injection of 110 mg STZ/kg b.w.) and dexamethasone (DEX)-treated rats (2.19 mg DEX/kg b.w. per day, given via drinking water, for 4 days).

When compared to appropriate controls, the effect of insulin on muscle protein synthesis in vitro was decreased by 40 and 60 % in DEX-treated and STZ-diabetic rats, respectively. In the presence of rapamycin, insulin-stimulated protein synthesis was diminished but not abolished, suggesting that insulin signalling in muscle involves both rapamycin-sensitive and rapamycin-insensitive pathways. The relative contribution of each pathway was not modified by DEX (rapamycin reduced insulin action by 33 and 37 % in control and DEX-treated rats, respectively). Rapamycin-dependent mechanisms were relatively more important in STZ rats (insulin action was reduced by rapamycin by 36 and 68 % in control and diabetic rats, respectively). The $p70^{S6K}$ protein level and basal $p70^{S6K}$ activity were not altered in any model. Whereas insulin caused an activation of $p70^{S6K}$ in STZ-diabetic rats, no stimulatory effect was observed in muscles of DEX-treated rats. On the basis of the present results we concluded that, even though an alteration of rapamycin-dependent elements occurs, the rapamycin-sensitive pathway does not account for the impairment of muscle protein synthesis observed in the case of DEX-induced or diabetes-associated insulin resistance.

Table I. P/WB (plasma/whole-blood) amino acid concentrations in carotid artery, portal and hepatic veins. Amino acids for which P/WB ratio is not different from 1 are not presented.

	Artery	Portal vein	Hepatic vein
Lys	$0.51^{a} \pm 0.02$	$0.58^{b} \pm 0.02$	$0.56^{b} \pm 0.02$
Thr	0.91 ± 0.02	0.90 ± 0.04	0.92 ± 0.01
Cys	1.27 ± 0.03	1.31 ± 0.05	1.31 ± 0.05
Gly	0.69 ± 0.005	0.69 ± 0.01	0.68 ± 0.01
Glu	$0.66^{a} \pm 0.08$	$0.64^{a} \pm 0.09$	$0.87^{b} \pm 0.06$
Gln	1.72 ± 0.09	1.66 ± 0.1	1.68 ± 0.1
Asp	0.36 ± 0.05	0.42 ± 0.05	0.38 ± 0.04
Asn	1.8 ± 0.11	1.76 ± 0.17	1.54 ± 0.25
Arg	1.31 ± 0.03	1.31 ± 0.05	1.32 ± 0.03

Ratios on the same line with different superscripts are significantly different ($P < 0.05$).

Communication no. 32

In vivo plasma and whole blood amino acid exchanges across the liver in pig.
N. Le Floc'h, N. Mézière, B. Sève (Station de recherches porcines, Inra, 35590 Saint-Gilles, France)

Amino acid metabolism studies have often been restricted to changes in the concentrations or movements of amino acids in the plasma compartment. Nevertheless, some authors have demonstrated that erythrocytes contribute to inter-organ amino acid exchanges (Elwyn et al., Am. J. Physiol. 222 (1972) 1333–1342; Houlier et al., Reprod. Nutr. Dev. 31 (1991) 399–410; Lobley et al., Br. J. Nutr. 75 (1996) 217–235). In pigs, there are no data available regarding the respective contributions of erythrocytes and plasma to the amino acid transport across the portal drained viscera.

Plasma and whole blood amino acid exchanges across the liver were examined in three hourly fed Large-White pigs (30.5 kg, mean live weight) which were surgically prepared with chronic insertion of catheters into four vessels: a mesenteric vein, the portal vein, an hepatic vein and the carotid artery. Blood flow was measured during 6 h of continuous PAH infusion through the mesenteric catheter. Whole blood, plasma amino acid and PAH concentrations were determined in the carotid artery and in portal and hepatic veins.

Lys, thr, gly, glu, asp and orn were present at higher concentrations in whole-blood than in plasma, whereas cys, gln, asn and arg were more abundant in plasma than in whole blood (*table I*). Except for lys and glu, the plasma/whole blood (P/WB) ratios showed no significant difference in the diverse vessels. The lysine P/WB ratio was significantly higher in the portal than in the artery suggesting that absorbed lysine was preferentially transported in the plasma. The glu P/WB ratio was higher in the hepatic vein suggesting that glu released from the liver was mainly transported in the plasma. The liver balance showed negative values for all amino acids except for glu, which exhibited a positive balance.

These data show that the contribution of plasma and whole blood to amino acid transport can be different according to the amino acids and the tissue under consideration. As a consequence, attention should be paid to the choice of blood sampling sites during amino acid metabolic studies.