Table I. (S Halimi *et al*)

	(Cer) n = *8*	*(Fr 58)* n = *8*	*(Fr58 + Met)* n = *8*
Ra mg/min	2.3 ± 0.1	2.5 ± 0.1	2.1 ± 0.6
Rd mg/min	7.7 ± 1.1*	3.3 ± 0.05*	6.3 ± 1.0**
HGP mg/min	–0.1 ± 0.3*	1.3 ± 0.2*	0.6 ± 0.3**

* $P \leq 0.5$; ** $P \leq 0.05$.

plus a 2-week treatment with metformin 200 mg/kg/d (Fr58 + Met). The euglycemic hyperinsulinic clamp method (2 mg/min/kg) was used in conscious animals. The basal appearance rate of glucose (Ra), the disappearance rate of glucose stimulated by insulin (Rd) and the hepatic glucose production (HGP) were measured by (^{3}H-3-D) glucose infusion (table I).

This study demonstrates that a high fructose diet administered in the early post-weaning period induces a huge insulin resistance in both hepatic and peripheral tissues. After a 2-week treatment with metformin, the fructose-induced increments in insulin resistance were reduced by 80 and 100% in peripheral and hepatic tissues. Thus, the high fructose diet in post-weaning rats is an excellent and useful experimental model of nutritional insulin resistance.

Effect of the nutritional state on insulin receptors and phosphoenolpyruvate carboxykinase in chicken kidneys. S Bisbis, M Derouet, J Simon *(INRA, Endocrinologie du Métabolisme et de la Croissance, Station de Recherches Avicoles, 37380 Nouzilly, France)*

In chickens, kidneys contribute to gluconeogenesis to a large extent during fasting (30%). Kidney PEPCK (phosphoenolpyruvate carboxykinase, the rate-limiting enzyme of gluconeogenesis) is under hormonal control, in contrast to liver PEPCK. This difference is due to the intracellular location of the enzyme: mitochondrial in liver (at least 95% of total activity) and partially cytosolic (20–45%) in kidneys. Insulin probably contributes to the control of kidney PEPCK. In the present study PEPCK activity was studied and insulin receptors were characterized in the kidneys of chickens submitted to the following nutritional conditions: fed state, 48-h fasted state, and 24-h refed state following a 48-h fast. PECK activity was increased by the 48-h fast and returned to normal after refeeding (comparisons made with the *ad libitum* fed state). Binding of ^{125}I-insulin to the chicken kidney membranes was time-, temperature- and protein-dependent and specific: unlabelled insulin was more potent than IGF-1 at inhibiting tracer binding (about 6 times). Insulin binding was significantly higher ($P < 0.05$) following the 48-h fast and lower ($P < 0.05$) following refeeding compared to *ad libitum* feeding. Since receptor affinity (as estimated by insulin concentration inhibiting tracer binding by 50%) was similar irrespective of the nutritional state (6–8 ng/ml), the changes in insulin binding are explained by changes in membrane insulin receptor number. The sizes of α and β subunits of kidney receptors were similar to those of liver receptors: 135 and 90 kDa, respectively. Following Triton X-100 solubilization of membranes and wheat germ agglutinin purification, insulin-stimulated autophosphorylation of the β-subunit was decreased non-significantly by fasting. Phosphorylation of the artificial substrate, poly (Glu-Tyr) 4:1, was significantly decreased by the 48-h fast at high insulin concentrations (10 and 100 nM). In conclusion, typical insulin receptors are present in chicken kidneys, the number of receptors is inversely correlated to plasma insulin levels, and the kinase activity of receptors is inhibited by prolonged fasting. This suggests that insulin action in chicken kidneys is controlled by several steps at the receptor level.

Regulation of GLP-1 release: study with a model of isolated vascularly perfused rat colon. P Plaisancié [1,2], C Bernard [1], JA Chayvialle [1], JC Cuber [1,2] *([1] INSERM Unité 45, Hôpital É-Herriot, Pavillon Hbis, 69437 Lyon Cedex 03; [2] INRA, Unité d'Ecologie et de Physiologie du Système Digestif, 78350 Jouy-en-Josas, France*)

Glucagon-like peptide-1 (GLP-1), a potent insulinotropic peptide, is produced with peptide YY in open-type endocrine L-cells localized in the ileum and the colon. These cells can receive various chemical stimuli from the intestinal chyme at their apical side and neural and blood-borne stimuli at their basolateral side. The mechanisms

that govern GLP-1 secretion are poorly know. To investigate the secretory activity of the GLP-1 producing cells, a model of isolated vascularly perfused rat colon was developed. In this preparation, polarized endocrine cells are submitted to well-defined luminal and blood-borne stimuli.

Loops of proximal colon were separated from adjacent tissues and perfused *via* the superior mesenteric artery with a Krebs–Heinseleit buffer containing 25% erythrocytes, 3% bovine albumin, amino acids and glucose. After a 20-min control basal period, stimuli were applied for 30 min. GLP-1 was measured in the portal effluent with antiserum 199D that cross-reacts 100% with GLP-1 (7–36 amide), 84% with GLP-1 (1–36 amide) and less than 0.1% with other peptides of the glucagon family. Luminal infusion of glucose (5%) induced GLP-1 release (plateau at 45 ±10 fmol/2 min from a basal value of 16 ± 2 fmol/2 min, $p < 0.05$) while starch (0.5%), oleic acid (100 mM), amino acids (29.6 g/l) and short-chain fatty acids (acetate, propionate, butyrate: 5, 20, 100 mM) did not modify basal secretion. Pectin (0.1–2%) produced GLP-1 secretion (maximal value at 200% above basal with 0.5% pectin). Cellulose or gum arabic did not release GLP-1. Hyodeoxycholate, the major bile acid in the colon (2–20 mM), induced a significant GLP-1 release (secretion rate 35–46 fmol/2min, $p < 0.05$) while cholate or deoxycholate did not. Arterial infusion of the neuropeptide bombesin (10^{-7} M) produced a biphasic GLP—1 secretion (transient rise at 290% of basal followed by a sustained response: 400% of basal). Calcitonin gene-related peptide (5 x 10^{-8} M), isoproterenol (β-adrenergic agonist, 10^{-6} M) and bethanechol (cholinergic agonist, 10^{-4} M) induced a sharp GLP-1 release. The effects of 3 hormones released by the proximal gut on the secretion of GLP-1 were then tested. The glucose-dependent insulinotropic polypeptide: GIP (0.25, 0.5, 1 nM) provoked a dose-dependent GLP-1 response (integrated release: RI of 71 ± 48 fmol/30 min at the physiological concentration of 0.25 nM, NS: maximal effect at the supraphysiological concentration of 1 nM, RI: 410 ± 71 fmol/30 min, $P < 0.001$). CCK (100 pM) and secretin (50 pM) had no effect on GLP-1 release. Finally, the combined perfusion of GIP (0.25 or 1 nM, arterial) and butyrate (5 or 20 mM, luminal) produced a GLP-1 secretion that was 4–6 fold higher than the sum of individual responses.

In conclusion, GLP-1 release is induced by luminal factors, neurotransmitters and an intestinal hormone (GIP). The synergistic effect between butyrate and GIP suggest that GLP-1 secretion is the result of complex relationships between factors operating at the luminal and basal sides of L cells in the colon.

IV. Energy expenditure

Determination of energy cost of standing in preruminant calves from continuous measurements in respiration chambers.

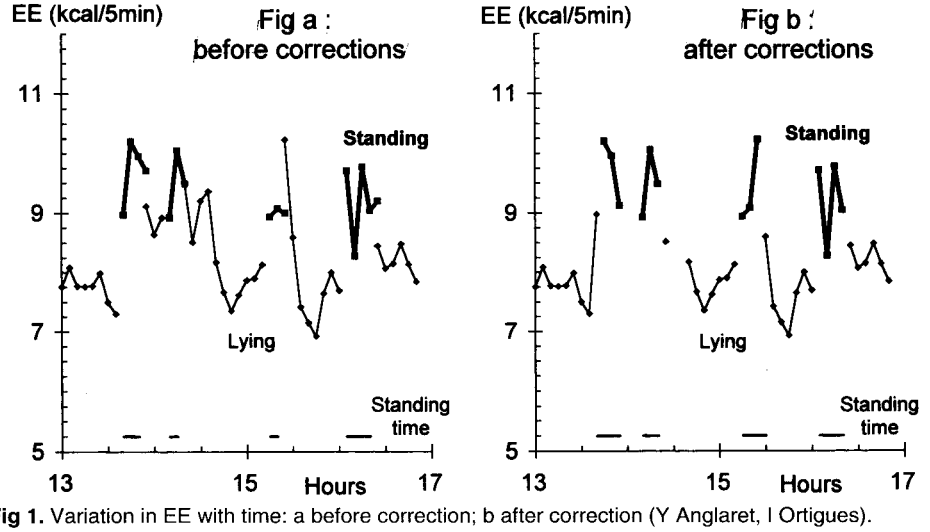

Fig 1. Variation in EE with time: a before correction; b after correction (Y Anglaret, I Ortigues).