enzyme appeared practically inactive in the same conditions (Table 1). This different inhibitory effect of pyrazole in the two species was also observed with a concentration of 200 μM (Table 1), which produced a partial inhibition of the human enzyme and complete inhibition of the rat enzyme. These findings demonstrate that human, but not rat, liver contains a pyrazole-insensitive activity. The presence of pyrazole-insensitive alcohol dehydrogenase activity in human liver has been reported in another study (Li *et al.*, 1977), and, since it exhibits a high K_m for ethanol, it may influence the ethanol-elimination rates *in vivo* when blood ethanol concentrations are very high. Present results indicate that this pyrazole-insensitive activity is absent from rat liver. At the same time, the lower enzyme activity and K_m and $V_{max.}$ values found for rat liver alcohol dehydrogenase indicate that in this species the enzyme system becomes saturated at lower ethanol concentrations than in humans, which would favour either its greater elimination in unoxidized form or its greater transformation through the alternate pathway, e.g. the microsomal ethanol-oxidizing system (Orme-Johnson & Ziegler, 1965) and/or catalase (De Duve & Baudhuin, 1966). This question is as yet unresolved, and the species-specific effects described should be considered when using the rat as an experimental model for alcohol effects in humans.

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Liver glycogen and glyceride glycerol formation as compared with glucose synthesis in 24h-starved virgin and pregnant rats

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Enhanced liver glucose production during starvation is initially sustained by augmented glycogenolysis, which causes a rapid decrease in glycogen stores (Herrera & Freinkel, 1968; Phan *et al.*, 1974). An increase in liver gluconeogenesis also occurs soon after the onset of starvation (Schimmel & Knobil, 1970; Llobera & Herrera, 1980), not only contributing to the net glucose release by the liver but avoiding complete depletion of its glycogen content. In this way, liver glycogen synthesis in starvation represents a gluconeogenic process. A similar mechanism seems to occur in the synthesis of glyceride glycerol from gluconeogenic substrates, which is also enhanced with starvation (Soley *et al.*, 1983). To determine whether there is a constant relation between the synthesis of either liver glycogen or glyceride glycerol and glucose production independent of the gluconeogenic substrate used, in the present work we administered intravenously 1 μCi of [U-^{14}C]alanine, [3-^{14}C]pyruvate or [U-^{14}C]glycerol (from The Radiochemical Centre, Amersham, Bucks., U.K.) containing 0.2mmol/200g body wt. to 24h-starved virgin rats. As gluconeogenesis is known to be greatly increased during starvation at late gestation in the rat (Herrera *et al.*, 1969), the same experiment was performed in 24h-starved 21-day pregnant rats, to determine also whether differences in the gluconeogenetic activity alter such relations. Animals were decapitated 10min after injection and blood [^{14}C]glucose, liver [^{14}C]glycogen and [^{14}C]glyceride glycerol were purified and determined as described elsewhere (Herrera *et al.*, 1969; Chaves & Herrera, 1980; Carmaniu & Herrera, 1980). As shown in Table 1, the ratio of liver [^{14}C]glycogen to blood [^{14}C]glucose, corrected to 200g body wt., was the same with all three substrates and in both pregnant and virgin animals. The ratio of liver [^{14}C]glyceride glycerol to blood [^{14}C]glucose was always higher than that of [^{14}C]glycogen to [^{14}C]glucose and, although values did not differ among substrates, they were consistently higher for pregnant than for virgin animals.

The present results show that in the starved condition there is a constancy in the proportional amount of newly synthesized glucose which is converted into liver glycogen or released to

Table 1. *Ratios of liver [^{14}C]glycogen or [^{14}C]glyceride glycerol to circulating [^{14}C]glucose synthesized from gluconeogenic substrates in the 24h-starved virgin or 21-day-pregnant rat*

Rats were intravenously injected with 1 μCi of [U-^{14}C]alanine, [3-^{14}C]pyruvate or [U-^{14}C]glycerol containing 0.2mmol/200g body wt. and killed 10min thereafter. Determinations were performed as previously described (Herrera *et al.*, 1969; Chaves & Herrera, 1980; Carmaniu & Herrera, 1980) and ratios were calculated for individual animals and expressed as means ± S.E.M. for five to ten rats per group. *P* refers to the statistical comparison between virgin and pregnant animals for each ^{14}C-labelled substrate (N.S. = not significant, $P > 0.05$). Statistical comparisons between values from different substrates were never significant ($P > 0.05$).

^{14}C-labelled substrate Rats ...	Liver [^{14}C]glycogen/blood [^{14}C]glucose			Liver [^{14}C]glyceride glycerol/blood [^{14}C]glucose		
	Virgin	Pregnant	*P*	Virgin	Pregnant	*P*
L-[U-^{14}C]Alanine	0.44 ± 0.05	0.54 ± 0.09	NS	2.33 ± 0.45	6.99 ± 0.59	<0.001
[3-^{14}C]Pyruvate	0.55 ± 0.07	0.62 ± 0.05	NS	2.64 ± 0.25	5.60 ± 0.41	<0.001
[U-^{14}C]Glycerol	0.42 ± 0.07	0.54 ± 0.13	NS	2.90 ± 0.31	7.57 ± 1.22	<0.01

circulation, and this process seems to be independent of the administered substrate. Since glycogen stores remain low in spite of parallel enhancement of liver glycogenolysis and gluconeogenesis with starvation, it may be proposed that in this condition a considerable proportion of gluconeogenic substrates is initially transformed to liver glycogen, which becomes a direct precursor of circulating glucose. Enzymic conditions for maintaining simultaneous enhancement of liver glycogenesis and glycogenolysis have been identified in the starved rat liver (Roach & Larner, 1976; Hems & Whitton, 1980). This persistent futile cycle in the presence of increased gluconeogenesis may have two main functions: (i) to ensure maintenance of a permanent minimum glycogen content in liver, which is required as primer, and (ii) to allow rapid recovery of glycogen stores whenever food is available after starvation while gluconeogenesis is still augmented.

Formation of glyceride glycerol from gluconeogenic substrates may be facilitated in the starved condition by the cytosolic redox state of the liver (Lindall & Lazarow, 1964; Williamson *et al.*, 1967; Berdanier *et al.*, 1979) facilitating esterification of incoming fatty acids. This hypothesis is supported by the augmented proportional formation of [^{14}C]-glyceride glycerol from the administration tracers in the starved late-pregnant rats (Table 1), where it is known that lipolysis is greatly enhanced (Leat & Ford, 1966; Knopp *et al.*, 1970), inducing the liver to increase its contribution of gluconeogenetic substrates for the synthesis of glyceride glycerol. This explanation also coincides with the fact that liver is the main receptor site of unesterified fatty acids released from adipose tissue by lipolysis (Mampel *et al.*, 1981), which is known to be enhanced in the starved condition (Schimmel & Knobil, 1970). Esterification of fatty acids in liver also changes proportionally according to fatty acids availability (Debeer *et al.*, 1981); thus deviation of gluconeogenic intermediates to glyceride glycerol formation in the starved condition accounts for the temporary esterification of the incoming fatty acids in the liver.

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Transfer from mother to foetus of L-alanine and glycerol in fed and 48h-starved pregnant rats

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Maternal malnutrition or starvation in the rat at late gestation causes intrauterine growth retardation (Rosso, 1977; Anderson *et al.*, 1980; Ahokas *et al.*, 1981). Although it has been hypothesized that decreased maternal–foetal nutrient transfer may occur during periods of food restriction, this has not been directly tested in the rat. Placental transfer in this species has only been examined indirectly by giving a radioactive substrate to the mother and determining its appearance in the foetus. With this method, the administered tracer is diluted in the maternal circulation before it becomes available to the placenta. Metabolic adaptation occurring in the late-pregnant rat is augmented in the starved condition (Freinkel, 1965), thus differences in the availability of substrates to the foetus when the mother is deprived of food may be an indirect consequence of changes in maternal metabolism, and/or an alteration in the actual placental transfer activity. To determine which of these possibilities is correct, in the present study our method (Lasunción *et al.*, 1983) for determination of the metabolite transfer from rat mother to her foetuses was used in fed and 48h-starved 21-day-pregnant rats. The method consists basically of a 20min infusion through the left uterine artery of a medium containing a ^{14}C-labelled metabolite. In this way, the left uterine horn received the tracer directly, whereas it reached the right horn after dilution in the mother's circulation. Therefore analysis of radioactivity present in foetal blood from both uterine sides and in the maternal circulation permits estimation of actual nutrient transfer.

In the present study, L-[U-^{14}C]alanine and [U-^{14}C]glycerol were used as tracers, since they cross the placenta by different mechanisms: the first by active transport against a gradient (Hill & Young, 1973), and the second by diffusion in favour of the gradient (Gilbert, 1977). It is also known that circulating concentrations of these maternal metabolites are affected conversely by starvation: L-alanine decreases (Girard *et al.*, 1977), glycerol increases (Girard *et al.*, 1977). As shown in Table 1, 20min after infusion of L-[U-^{14}C]alanine through the left uterine artery, total radioactivity in plasma of foetuses from the left uterine horn was much greater than in their mothers, whereas radioactivity in plasma of foetuses from the right side was significantly lower than in the mother's plasma or in foetuses from the left side. These results confirm previous findings (Hill & Young, 1973) and indicate that, whereas plasma radioactivity in foetuses from the left side corresponds to the direct transfer of L-[U-^{14}C]alanine infused through the left uterine artery, plasma radioactivity present in the foetuses from the right side results from dilution of the tracer in the maternal circulation. When the same experiment was conducted in pregnant rats after 48h of starvation, the radioactivity present in plasma of foetuses from the left uterine horn did not differ from that in foetuses from the same side in fed mothers (Table 1). There was, however, a significant increase in total plasma radioactivity in foetuses from the right side in starved as compared with fed mothers. This difference parallels the increased plasma radioactivity in the 48h-starved as compared with the fed mothers (Table 1); thus the ratio of plasma radioactivity in mothers and their foetuses remained unchanged in both fed and starved conditions. The present results indicate that maternal-foetal transfer of L-alanine is not affected by starvation, and any change in the availability of this amino acid