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Utero-placental transfer and fetal utilization of β -hydroxybutyrate in swine

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UTERO-PLACENTAL TRANSFER AND FETAL UTILIZATION OF β -HYDROXYBUTYRATE IN SWINE

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Summary

Utero-placental transfer of β -hydroxybutyrate (BOHB) in swine was investigated during late gestation using three primiparous sows. There were no measureable artery-vein differences for BOHB concentration in whole blood and concentrations were low in both the sows and fetuses. Radioactive BOHB rapidly crossed the utero-placental unit and was incorporated into the liver, adipose, lung, and heart tissue (3540, 3674, 1214, and 528 dpm/g wet weight, respectively). The largest amount of radioactivity for all tissues was found in the phospholipid fraction. These data demonstrate that the swine utero-placental unit is permeable to BOHB and indicate that this ketone body is used by the fetus for synthesis of fatty acids and cholesterol for both structural components and energy stores.

Introduction

Previous research has shown that inducing maternal, nutritional ketosis in swine during late gestation alters enzyme activity and increases hepatic acetate and BOHB utilization for lipogenesis in the fetus, resulting in improved neonatal pig survival. This suggests that altered fetal development caused by maternal ketosis may result in a glucose-sparing effect, making additional glucose available within the fetus for glycogen synthesis. In addition, this may also provide a metabolic setting that favors fat synthesis in the fetus. By increasing fetal storage of glycogen and fat, the neonatal pig may have increased amounts of energy storage for improved survival.

Therefore, the purpose of this study was to determine if BOHB crosses the swine uterus and placenta and to determine if it is utilized by the fetus during late gestation in sows fed a corn-soybean meal diet.

Experimental Procedure

Three primiparous sows (Yorkshire x Duroc) were assigned to surgery on either d 108 or 112 of gestation, with feed being withheld overnight prior to surgery. The sows were anesthetized (thiamylal sodium and methoxyflurane) and an injection catheter inserted into the abdominal aorta (via a femoral artery), such that the tip resided 28 cm anterior to the bifurcation of the internal iliac.

D(-)-3-hydroxy[3- 14 C] butyric acid¹ (sodium salt, 58 mCi/mmol) was purchased from a commercial source and utilized without further purification. Immediately before use, the [14 C] β -hydroxybutyrate was mixed with 5 ml of sterile

¹Amersham Corp., Arlington Heights, IL.

.90% NaCl (w/v) and each sow injected with 200 or 250 μCi of [^{14}C]-hydroxybutyrate.

The pregnant uterus was exposed through a paramedial incision and the umbilical cord of each fetus exposed sequentially. Blood samples were collected simultaneously from the uterine artery (UA), uterine vein (UV), umbilical artery (FA), and umbilical vein (FV) at 2.5 and 5 minutes and every 5 minutes thereafter until 50 minutes post-injection. Blood samples were immediately deproteinized, centrifuged, and the supernatant stored at -20°C until assayed.

Blood samples were assayed for BOHB (enzymatically), with BOHB isolated for specific activity by high-pressure liquid chromatography and radioactivity determined by a liquid scintillation spectrophotometer. In addition, separate blood and tissue samples were extracted and the major lipid fractions separated by thin-layer chromatography (TLC) to determine destination of the labelled BOHB.

Results and Discussion

Mean concentrations of BOHB assayed throughout the experiments are shown in Table 1. Under conditions of adequate nutrition, concentrations of BOHB were very low in sows and fetuses in these experiments, making it difficult to determine net uptake by the fetus.

Specific activity for BOHB is summarized in Figures 1, 2, and 3 for the three experiments. Specific activities determined for UA and UV were high between 2 and 5 minutes after injection but declined rapidly during the experiments. The average decrease in specific activity between FV and FA ranged from 22.5% to 32.8% in the three experiments.

Incorporation of radioactivity into fetal tissue lipids during the initial 25 minutes following injection is shown in Table 2a. Results of radioactivity determined in fetal tissues at 5, 15, or 25 minutes were similar. Considerable amounts of radioactivity were present in the fetal tissue extracts. The largest amounts of total radioactivity (dpm/g wet weight) were found in the liver and adipose tissue, followed by the lung and heart tissues. There was a rapid incorporation of radioactivity into fetal tissues within 5 minutes. However, only trace amounts of radioactivity were found in the plasma lipid fractions of maternal and fetal blood after 5 to 10 minutes postinjection, suggesting that the radioactive label was associated with BOHB.

The distribution of the radioactivity incorporated into fetal tissues (Table 2b) was determined by subjecting lipid extracts to TLC. The phospholipid fraction contained the largest amount of radioactivity in each of the tissues analyzed. Liver tissue contained similar amounts of radioactivity in the cholesterol-diacylglycerol, FFA, and triacylglycerol fractions, whereas adipose tissue contained about 27 and 20% of the radioactivity in the triacylglycerol and cholesterol-diacylglycerol fractions, respectively. In lung and heart tissues, about 30% of the radioactivity was found in the cholesterol-diacylglycerol fraction. However, cholesteryl esters contained only trace amounts of activity in each of the tissues assayed. Similarly, radioactivity was low in the fatty acid fraction of adipose, lung, and heart tissue.

Table 1. Concentration of β -Hydroxybutyrate in Maternal and Fetal Blood.

	Uterine Vein	Uterine Artery	Umbilical Vein	Umbilical Artery
β -hydroxybutyrate, mmole/l blood ^a	.0189 $\pm$.0078	.0197 $\pm$.0092	.0054 $\pm$.0018	.0063 $\pm$.0014
^a $\pm$ SE				

Table 2. Incorporation of Radioactivity into Fetal Tissue Lipids Following Maternal Injection of [3-¹⁴C] β -Hydroxy-Butyrate.^{ab}

(a) Total incorporation:

Item	Tissue			
	Liver	Adipose	Lung	Heart
Radioactivity, dpm/g wet weight	3540 $\pm$ 336	3674 $\pm$ 296	1214 $\pm$ 69	528 $\pm$ 47

(b) Relative incorporation into lipid fractions (percent of total):

Tissue	Lipid Fraction				
	Phospholipid	Cholesterol + Diacylglycerol	Fatty Acid	Triacylglycerol	Cholesteryl Esters
Liver	43.9 $\pm$ 6.0	16.3 $\pm$ 4.2	19.7 $\pm$ 5.4	14.1 $\pm$ 6.5	6.0 $\pm$ 2.0
Adipose tissue	38.6 $\pm$ 1.4	20.9 $\pm$ 5.6	9.4 $\pm$ 3.6	29.0 $\pm$ 3.5	2.3 $\pm$ 2.1
Lung	57.7 $\pm$ 5.6	26.3 $\pm$ 6.0	9.4 $\pm$ 4.7	3.8 $\pm$ 3.0	2.9 $\pm$ 2.3
Heart	53.5 $\pm$ 8.4	31.3 $\pm$ 7.7	5.1 $\pm$ 3.8	3.8 $\pm$ 2.4	6.4 $\pm$ 4.0

^aMean $\pm$ standard deviation.^bEach mean represents 6 fetuses.

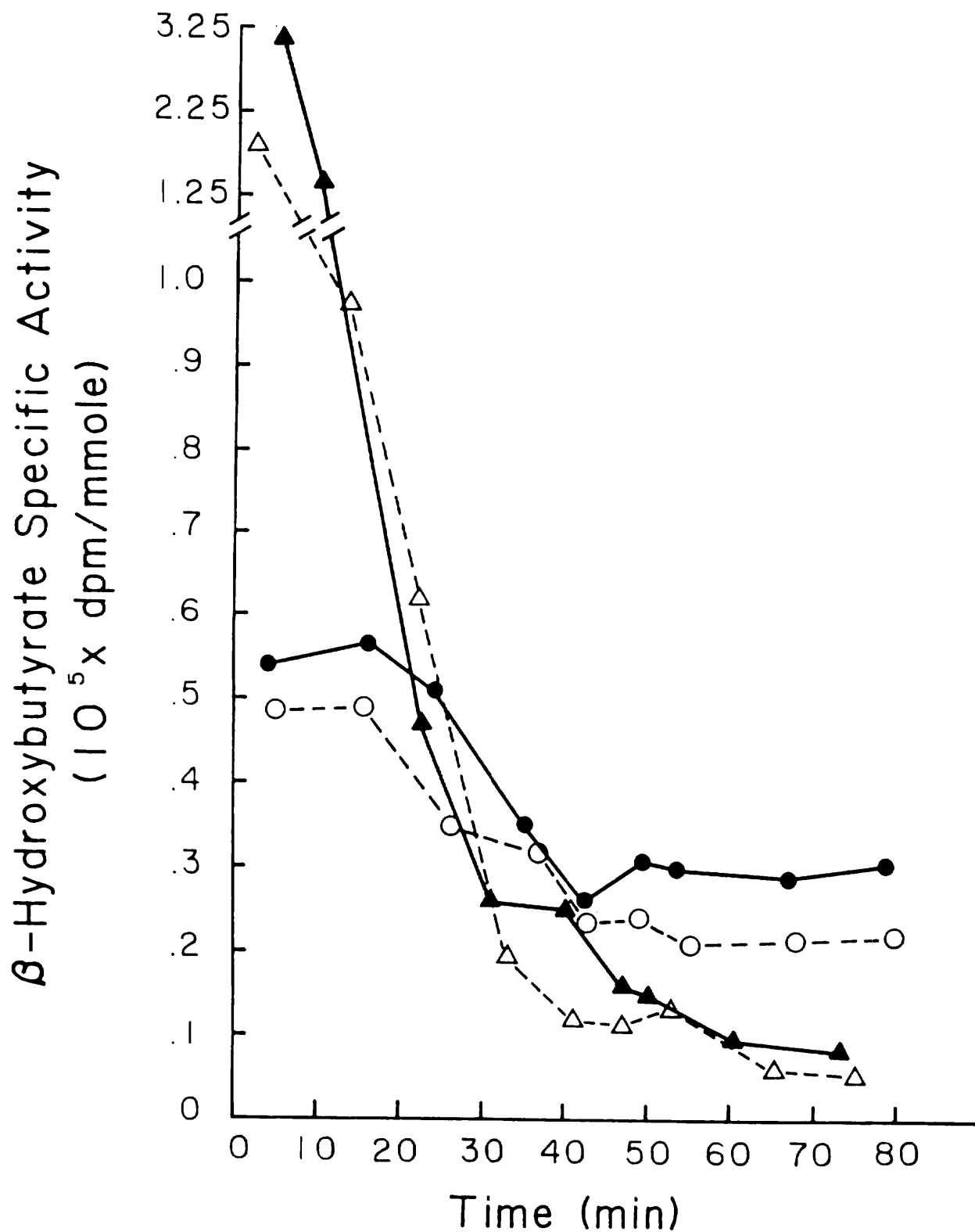


Figure 1. Specific activity of β -hydroxybutyrate in maternal and fetal blood following maternal injection of 200 μ Ci of [14 C] β -hydroxybutyrate (Exp. 1). Solid circle ($\bullet$) represents FV; open circle ($\circ$) represents FA; solid triangle ($\blacktriangle$) represents UV and open triangle ($\triangle$) represents UA.

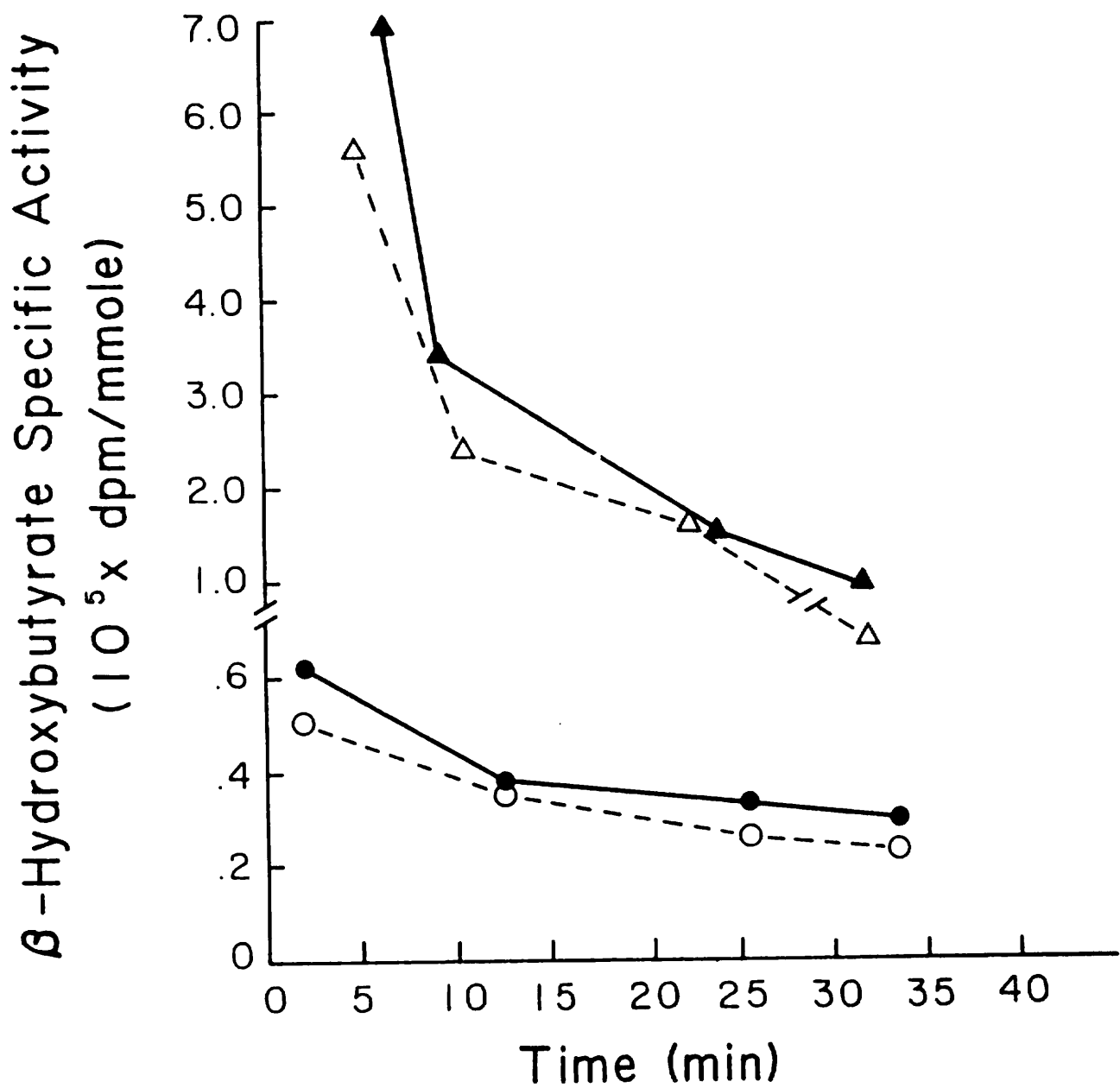


Figure 2. Specific activity of β -hydroxybutyrate in maternal and fetal blood following maternal injection of 250 μ Ci of [14 C] β -hydroxybutyrate (Exp. 2). Solid circle ($\bullet$) represents FV; open circle ($\circ$) represents FA; solid triangle ($\blacktriangle$) represents UV and open triangle ($\triangle$) represents UA.

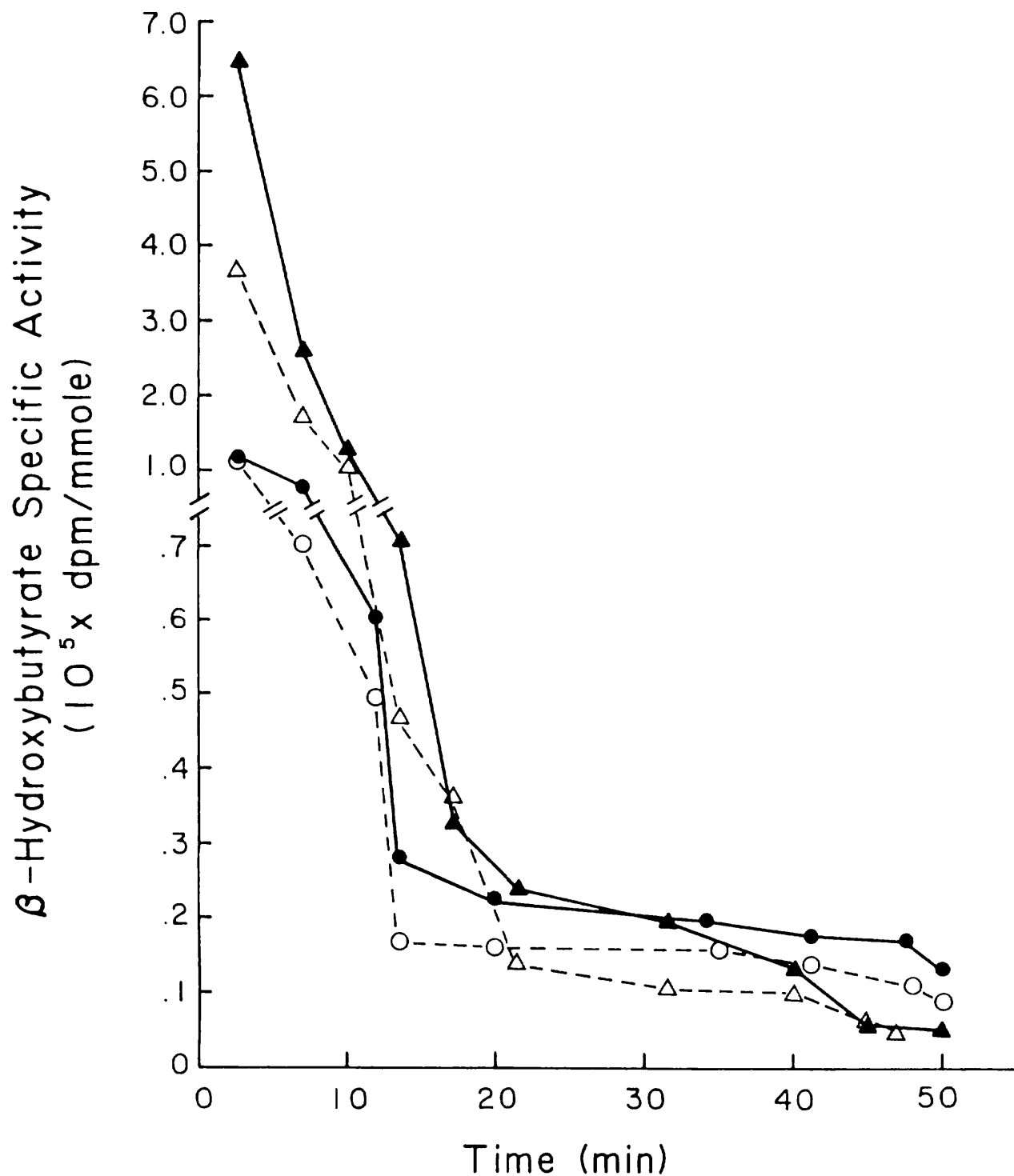


Figure 3. Specific activity of β -hydroxybutyrate in maternal and fetal blood following maternal injection of 250 μ Ci of [14 C] β -hydroxybutyrate (Exp. 3). Solid circle (●) represents FV; open circle (○) represents FA; solid triangle (▲) represents UV and open triangle (△) represents UA.