Tracer Experiments with Glucose

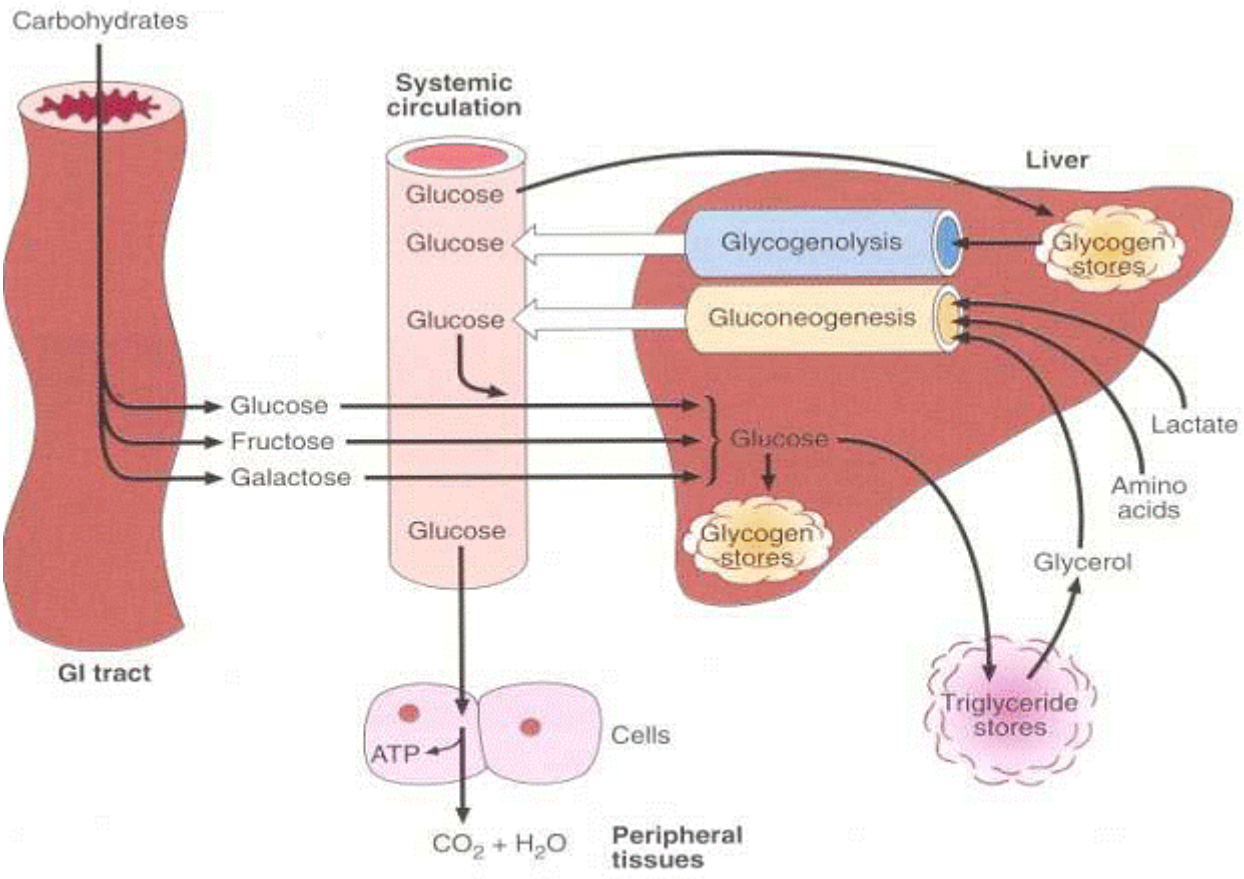

Fig. 1 **Glucose homeostasis.**

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Tracer Experiments with Glucose

Purpose

This publication will describe the technical details of using stable isotope tracers to determine glucose flux, recycling, oxidation, and synthesis rate from gluconeogenic precursors.

Introduction

The primed constant infusion of stable isotope labeled glucose has been extensively validated as a reliable approach to quantifying glucose kinetics. This approach can be used to explore whole body glucose homeostasis under various perturbations such as exogenous infusions of glucose and insulin. Sensitive methods for measuring labeled glucose in plasma have been developed to permit quantitation with 50 - 100 µl of plasma. Thus, these studies can be performed even in newborns.

Glucose Flux

Determines the whole body flux rate of glucose. If the amount of glucose intake is known, the glucose production rate can be calculated.

Protocol

Best Tracer: 6,6-D_2-Glucose
Priming Bolus Dose: 14.0 µmol/kg
Infusion Pump Speed: 0.174 cc/min
Infusion Rate: 11.5 µmol/kg/hr
Infusion Time: 140 min.
Sampling Times: 0, 90, 100, 110, 120 min. (Plasma)
Diet Protocol: Fasted or Fed
References: Bier et al., Diabetes **26**,1005-1015, 1977
Bier et al., Diabetes **26**, 1016-1023, 1977
Shaw and Wolfe, Surgery **97**,557-568, 1985

Glucose Recycling

Determines glucose and fructose recycling rates in addition to glucose flux and production rate. The measured rates of cycling in glycolysis and gluconeogenesis represents the total substrate cycling rates between glucose and glucose-6-phosphate and fructose-6-phosphate and fructose-1,6-diphosphate.

Protocol

Best Tracers: 6,6-D_2-Glucose and 2-D_1-Glucose
Priming Bolus Dose: 17.8 µmol/kg each tracer
Infusion Pump Speed: 0.174 cc/min
Infusion Rate: 13.2 µmol/kg/hr
Infusion Time: 140 min.
Sampling Times: 0, 90, 100, 110, 120, 130, 140 min. (Plasma)
Diet Protocol: Fasted or Fed
References: Shulman et al., J. Clin. Invest. **76**, 757-764, 1985
Miyoshi et al., J. Clin. Invest. **81**, 1545-1555, 1988

Glucose Oxidation

The rate of glucose oxidation can be calculated using a ^{13}C-labeled glucose tracer. Samples of blood are collected to determine the plateau level of ^{13}C-glucose in blood while expired breath samples are used to determine the enrichment of $^{13}CO_2$. The bicarbonate pool is primed with $NaH^{13}CO_3$.

Protocol

Best Tracer: U-($^{13}C_6$)-glucose
Priming Bolus Dose: 1.1 µmol/kg U-($^{13}C_6$)-glucose
Priming Bolus Dose: 0.6 mmol $NaH^{13}CO_3$
Infusion Pump Speed: 0.174 cc/min
Infusion Rate: 2.5 µmol/kg/hr
Infusion Time: 140 min.
Sampling Times: 0, 90, 100, 110, 120, 130, 140 min. (Plasma and Breath)
Diet Protocol: Fasted or Fed
References: Wolfe et al., Metabolism **28**, 210-219, 1979.
Robert et al., Diabetes **31**, 203-211, 1982.

Glucose Synthesis from Precursors

The rate of gluconeogenesis can be quantified from gluconeogenic substrates using ^{13}C stable isotope tracers. For example, to quantify the glucose-alanine relationship, one needs to determine the incorporation of alanine carbon into glucose. In addition, the fraction of lactate produced from alanine can be determined by measuring the enrichment of lactate. These studies require the infusion of [2,3-$^{13}C_2$]-alanine and 6,6-D_2-glucose as tracers.

Protocol

Best Tracers: [2,3-$^{13}C_2$]-alanine and 6,6-D_2-glucose
Priming Bolus Dose: 14 µmol/kg Each tracer
Infusion Pump Speed: 0.174 cc/min
Infusion Rate: 11.5 µmol/kg/hr Each tracer
Infusion Time: 140 min.
Sampling Times: 0, 90, 100, 110, 120, 130, 140 min. (Plasma and Breath)
Diet Protocol: Fasted or Fed
References: Kalhan et al., Metabolism **37**, 152-158, 1988.
