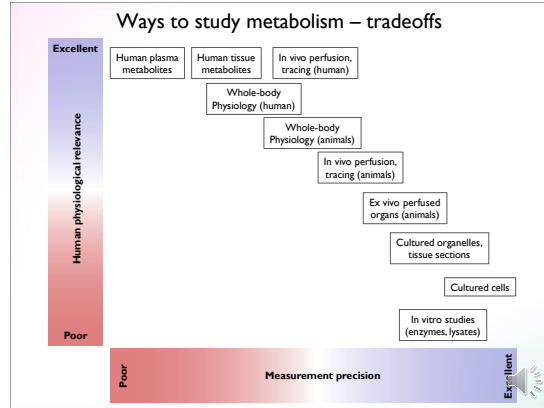



Course 2851 Principles of Metabolism
Metabolism and endocrinology programme, Karolinska Institutet

Lecture 17
Experimental considerations

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Culture systems

Batch culture

Organoids

Cells
HeLa

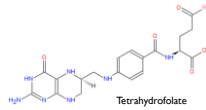
Tissue slices

Huch & Koo, Development 142:3113-3125, 2015.

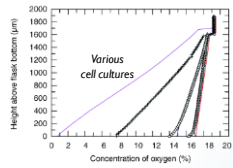
Mattei et al, PeerJ, 30:e932, 2015

Oxygen tension

- Low O₂ hinders respiration and some oxidases
- High O₂ oxidizes folates and thiol groups (also in proteins / enzymes)



O₂ profile in cell culture flask



Petersen et al, Cell Prolif 38:257-267, 2005.

What is "normoxia" ?

O₂ in vivo varies by tissue

1—10% saturation

Medium and batch culture effects

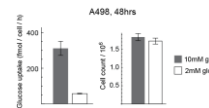
Culture media nutrient concentrations

Metabolite	Eagle's µM	DMEM µM	RPMI-1640 µM	Iscove's µM	Yessel's µM	William's µM
glucose	5,554	24,991	11,107	24,991	24,991	11,110
alanine	100			791	291	1,019
arginine	721	479	1,142	479	479	287
glutamate	101		137	513	513	340
glutamine	1,998	3,996	2,963	3,966	3,996	261
hydroxyproline			153			
proline	100		114	347	347	261
pyruvate	11	40	194	40	40	11
pyruvate	1,250	1,250		1,250	1,250	227

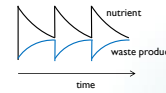
Vitamin deficiencies



Metabolism depends on nutrient concentrations

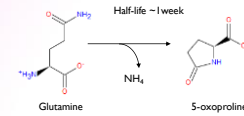


Medium changes



Various artifacts

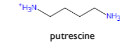
Glutamine degrades spontaneously



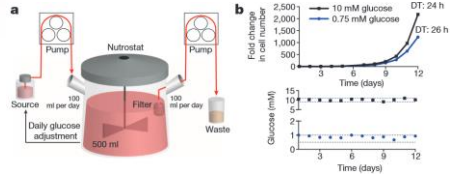
Serum can contain enzymes



Evaporation / volatile compounds

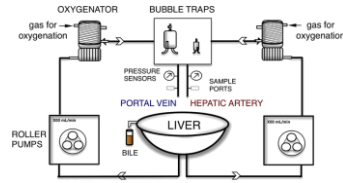


Continuous cell culture systems



Birsoy et al, Nature 508:108-112, 2014.

Ex vivo perfusion systems



Bruinsma et al, Scientific Reports 6: 22415, 2016.

Human plasma measurements

Data from venous blood (draining muscle)

Portals vein

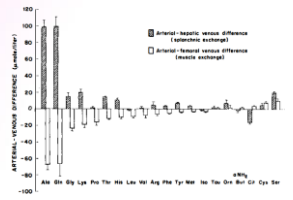
- Cephalic vein
- Right gastric vein
- Left, right portal vein
- Left, right gastric vein
- Splenic vein
- Gastro-epiploic vein
- Superior mesenteric vein
- Inferior mesenteric vein
- Veins from anterior/dorsal

Lymph drains into superior vena cava

Affected by various organs

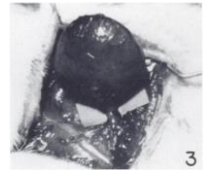
In vivo uptake / measurements

Artery - vein differences (human)



Felig, Ann Rev Biochem. 44:933-955, 1975.

Tumor model in vivo (rat)



Sauer, Cancer Res. 42:4090-97, 1982.