Attempts to generate liver organoids from rabbit iPSCs

Equipe de recherche

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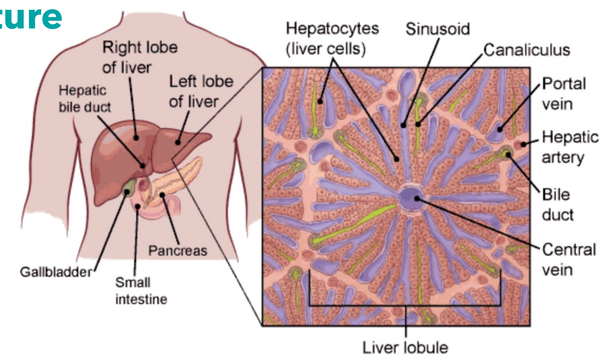
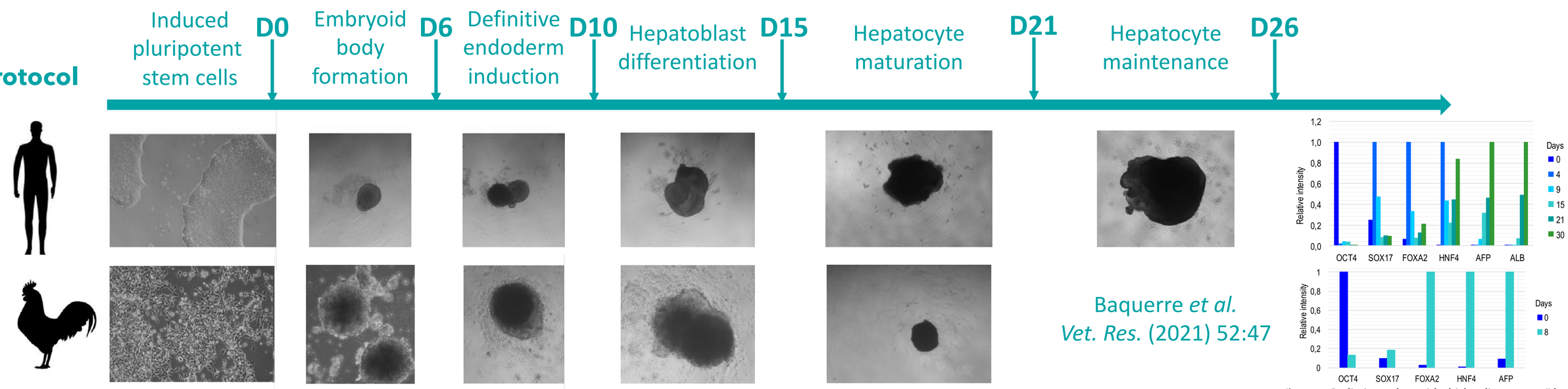
Introduction

The liver is a major and multifunctional organ responsible for the regulating of a multitude of complex metabolic processes. Despite the huge regenerative capacity of this organ, in humans, liver diseases are a major global health problem. The rabbit is often used as a model for non-alcoholic fatty liver disease or liver cancers, thanks to the similarities of its hepatic physiology and anatomy with those of humans. Nevertheless, the liver is a delicate organ in rabbit as well, due to its sensitivity to endogenous and exogenous toxins and to parasites responsible for coccidiosis and toxoplasmosis. The structure of the liver is composed of 80% of hepatocytes organized in lobules around blood vessels and bile capillaries composed of cholangiocytes. Hepatic organoids have been mainly produced in humans from healthy and diseased liver biopsies or from induced pluripotent stem cells (iPSCs). Different protocols are therefore available creating organoids of various complexity and function. However, very few examples are published in agronomic species. Our project aims at producing hepatic organoids from rabbit iPSCs, using a protocol that mimics the liver embryonic development.

Liver structure

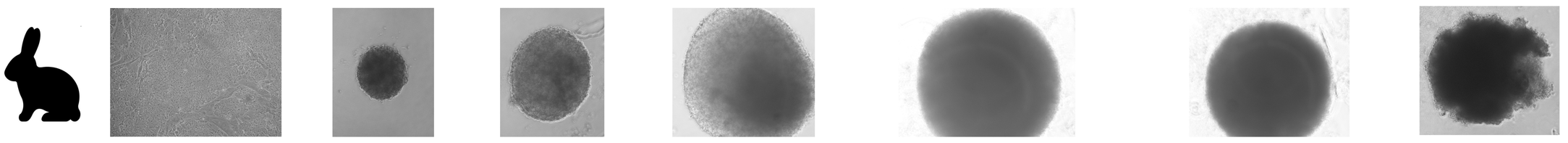


Protocol



Baquerre *et al.* *Vet. Res.* (2021) 52:47

Media



DMEM/F12
KOSR
FGF2
Feeder cells

DMEM/F12
KOSR
FGF2
Hydrogel

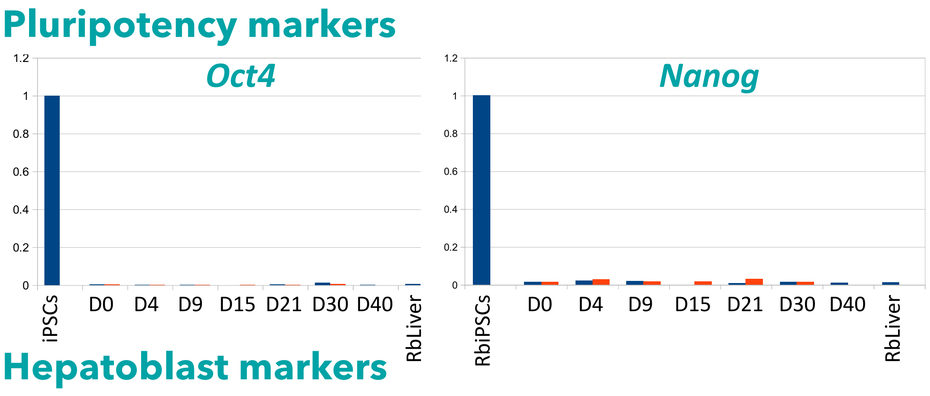
RPMI + B27
Wnt3a
Activin A
NaBu /Zebularin

RPMI + B27
FGF
Nicoinamide
Glutamax
NaBu /Zebularin

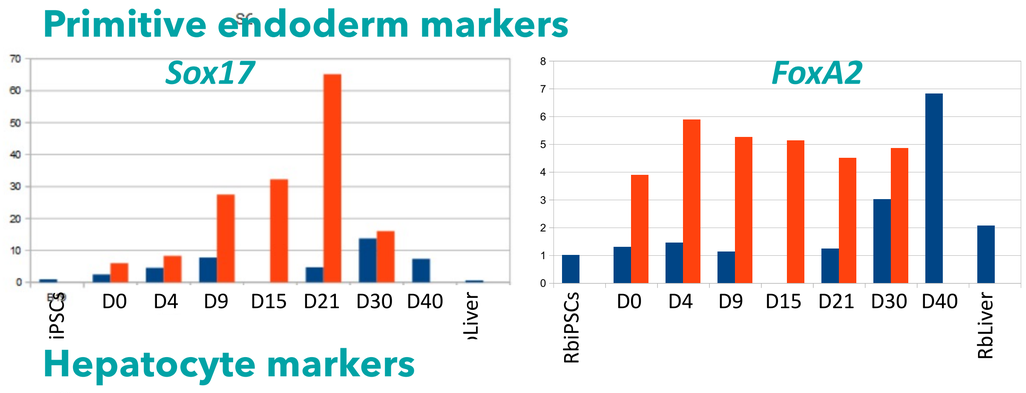
Hepatozyme
HGF
Oncostatin
Glutamax

William'E Medium
Bovin insulin
KOSR
BSA
NaPy
Hydrocortisone
HGF + FGF + VEGF + EGF

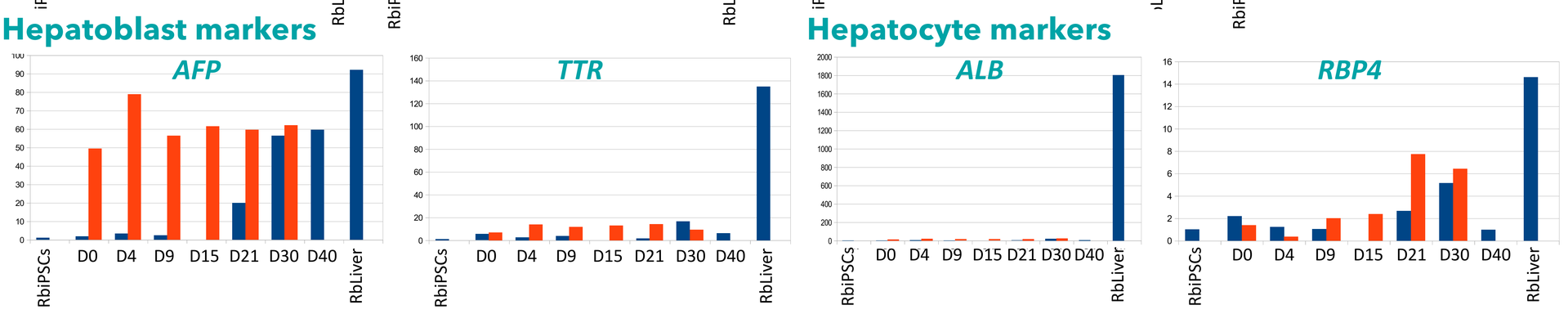
Pluripotency markers



Primitive endoderm markers



Hepatoblast markers



Hepatocyte markers

Conclusion

The use of a protocol defined for human cells with adapted media supplemented with Zebularin allows the assembly of liver organoids of fairly good quality from rabbit iPSCs. However, the hepatocyte medium needs to be improved since the expression of the albumin gene remains very low. Similarly, the hepatocyte maintenance medium needs to be carefully defined since the organoids dissociated after 21 days of culture.

Overall, these data are encouraging for the future creation of a 3D cultured rabbit liver model.

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