CLAIMS

What is claimed is:

1. A method of inducing formation of a liver organoid from definitive endoderm (DE), comprising the steps of:

a) contacting definitive endoderm (DE) with an FGF pathway activator and a GSK3 inhibitor, for a period of time sufficient to form posterior foregut spheroids; and

b) incubating said posterior foregut spheroids of step a) in the presence of retinoic acid (RA) for a period of time sufficient to form said liver organoid,

wherein the liver organoid is characterized by albumin secretion and comprises a luminal structure comprising internalized microvilli comprising mesenchymal cells, wherein said luminal structure is surrounded by polarized hepatocytes and basement membrane..

~~2. The method of claim 1, wherein the DE of step a) are not contacted with a BMP pathway inhibitor.~~

~~3. The method of claim 1 or 2, wherein the posterior foregut spheroids of step b) are not contacted with a BMP pathway inhibitor.~~

~~4~~2. The method of ~~any one of~~ claim~~s~~ 1~~-3~~, wherein the period of time of step a) is from about 1 day to about 3 days.

~~5~~3. The method of ~~any one of~~ claim~~s~~ 1~~-4~~ or 2, wherein the period of time of step b) is from about 1 day to about 5 days.

~~6~~4. The method of any one of claims 1~~-5~~3, further comprising c) incubating said liver organoid of step b) with hepatocyte culture medium for a period of time sufficient to mature said liver organoid.

~~7~~5. The method of claim ~~6~~4, wherein the period of time of step c) is from about 20 days to about 30 days.

~~8~~6. The method of claim ~~6~~ 4 or ~~7~~5, wherein the hepatocyte culture medium comprises at least one of hepatocyte growth factor, dexamethasone, or Oncostatin M.

~~9~~7. The method of any one of claims 1-~~8~~6, wherein the DE is derived from stem cells.

~~10~~8. The method of claim ~~9~~7, wherein said stem cells are human iPSCs.

~~11~~9. The method of any one of claims 1-~~10~~8, wherein said foregut spheroids are embedded in a basement membrane matrix.

~~12~~10. The method of claim ~~11~~9, wherein the basement membrane matrix is Matrigel.

~~13~~11. The method of any one of the preceding claims, wherein the liver organoid expresses alpha-fetoprotein (AFP), albumin (ALB), retinol binding protein (RBP4), cytokeratin 19 (CK19), hepatocyte nuclear factor 6 (HNF6), and cytochrome P450 3A4 (CYP3A4).

~~14~~12. The method of any one of the preceding claims, wherein the liver organoid has bile transport activity.