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A novel experimental model to understand cholangiocarcinogenesis

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AMMF

THE CHOLANGIOCARCINOMA CHARITY

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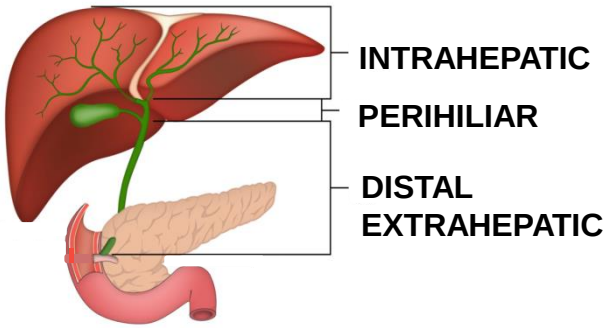
1. **Introduction**

2. **Models of cholangiocarcinogenesis**
    2.1. Deletion of JNK1/2 in NEMO mice

3. **Therapies against CCA**
    3.1. The use of CM272 in CCA

Facts about Cholangiocarcinoma



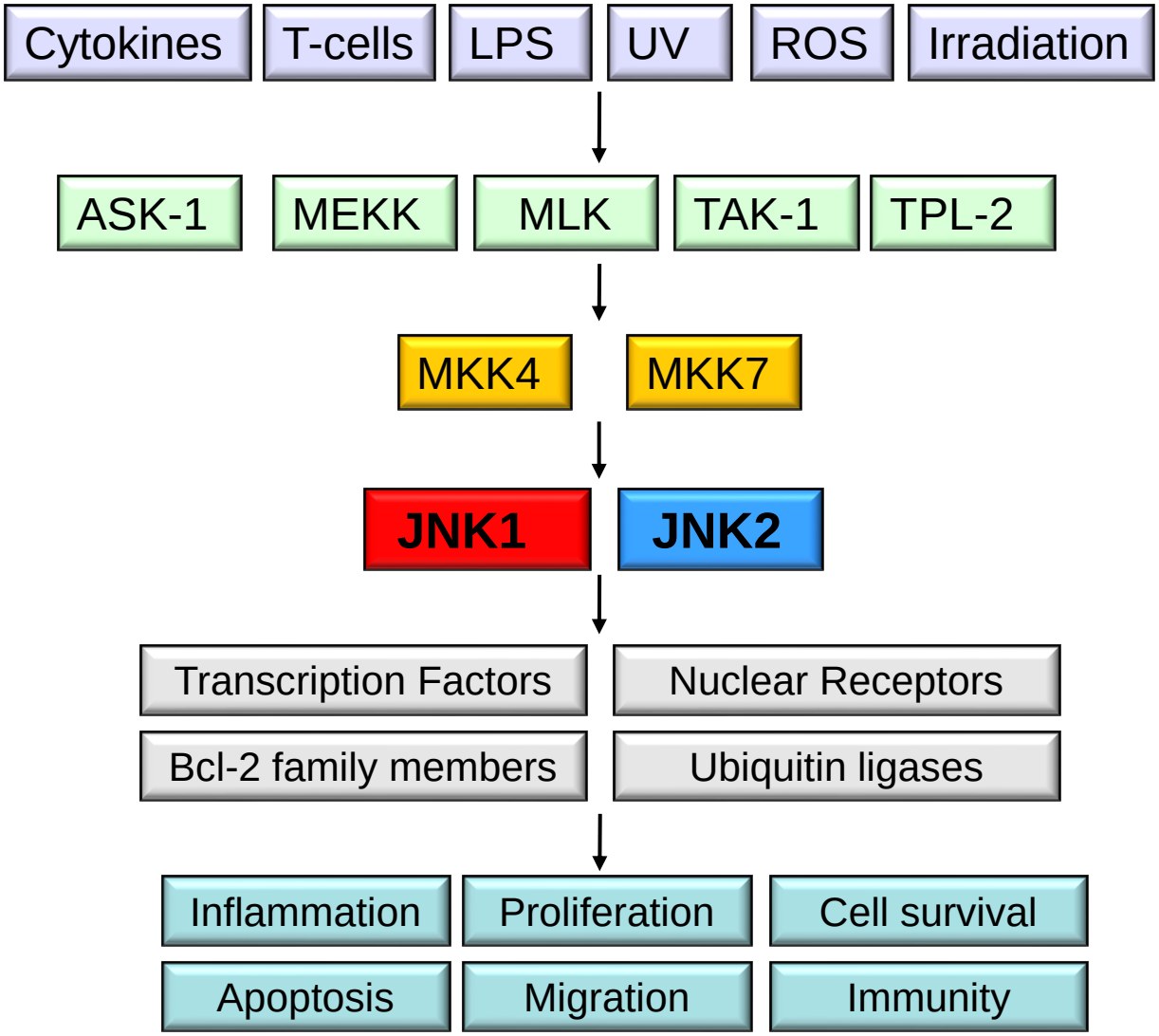
- Highly heterogeneus: important inter- and intratumour variability
- Incidence increasing
- Late diagnosis ➡ poor prognosis ➡ high mortality
- Current therapeutic strategies:
  Surgical resection
  Chemotherapy ➡ high CCA chemoresistance

- Numerous ***in vivo*** **and** ***in vitro*** **models**, as well as research with human samples, are helping to elucidate the main pathways implicated in CCA formation.

- As yet, however, **none** of these studies **recapitulate the human disease** and thus translation into improved patient outcome has not been achieved.

- The pathophysiology of CCA remains poorly understood. Therefore, there is an **urgent need for new models** to improve management of this insidious and devastating disease.

The mitogen-activated protein kinases (MAPK)



The c-Jun N-terminal kinases (JNK) pathway

- The c-Jun N-terminal kinases (JNK) are an evolutionarily conserved mitogen-activated protein kinase (MAPK) which, plays an important role in converting extracellular stimuli into a wide range of cellular responses, including **inflammatory** response, **stress** response, **differentiation**, and **survival**.

- Importantly, JNK has **lineage determinant functions** in liver parenchymal cells.

- In tumorigenesis, JNK can have an **anti- or pro-oncogenic** functions.

- Recently, it was reported that **JNK modulates CCA progression** by impeding JNK-mediating biliary proliferation.

- Altogether, these data indicate that JNK modulation would be of **therapeutic benefit** in CCA patients.

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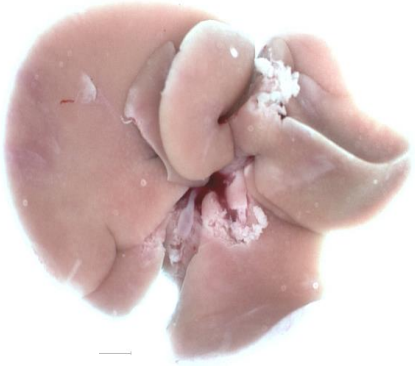
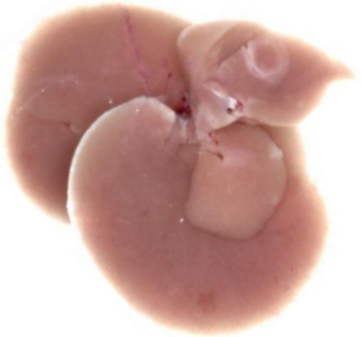
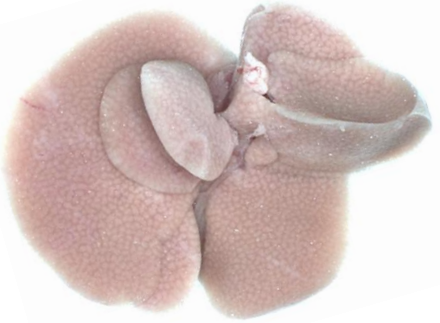
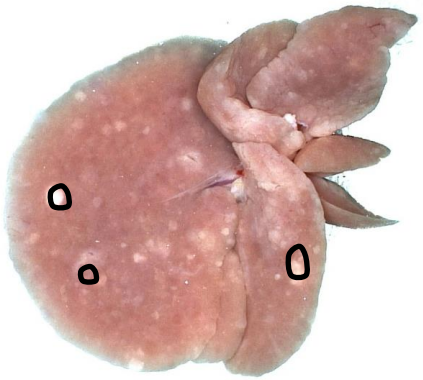
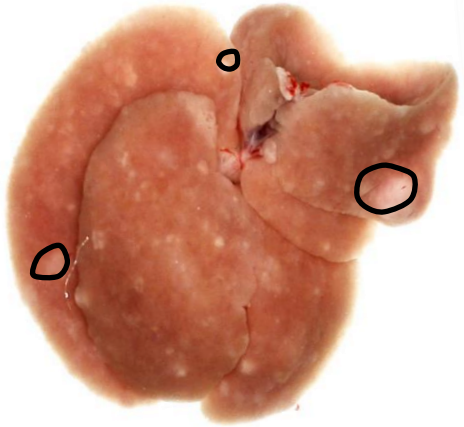
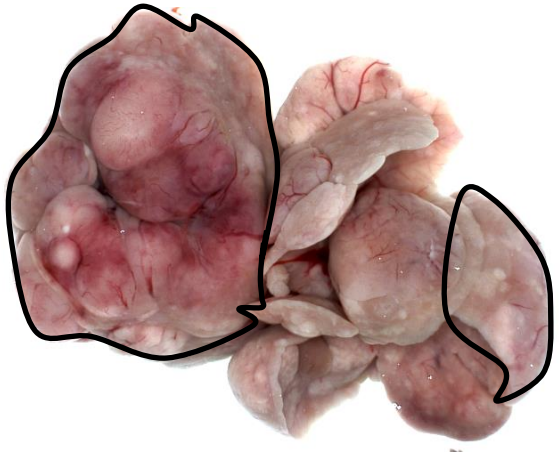
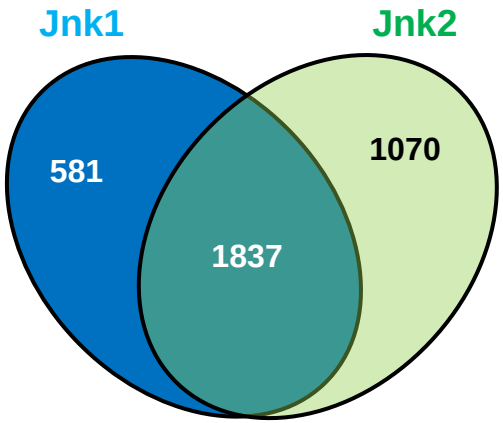
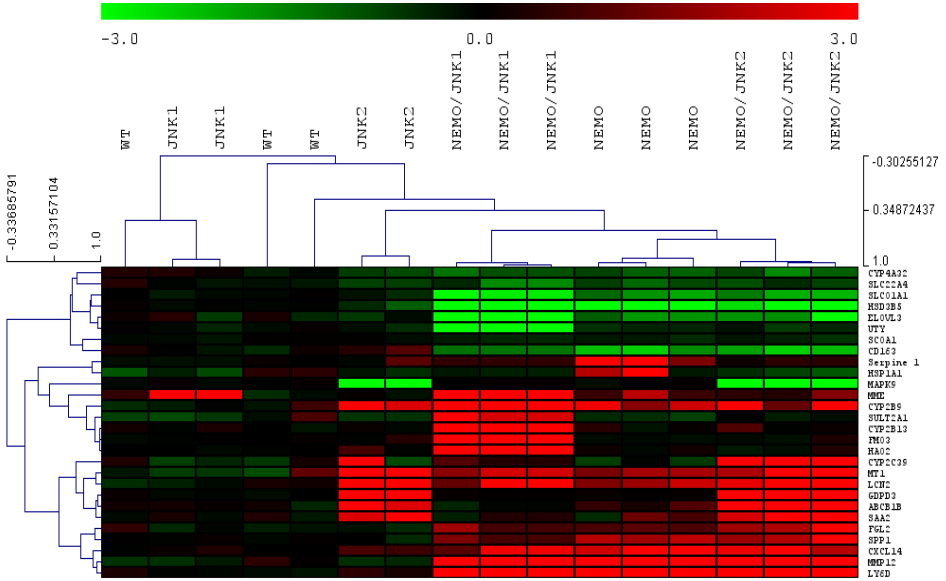
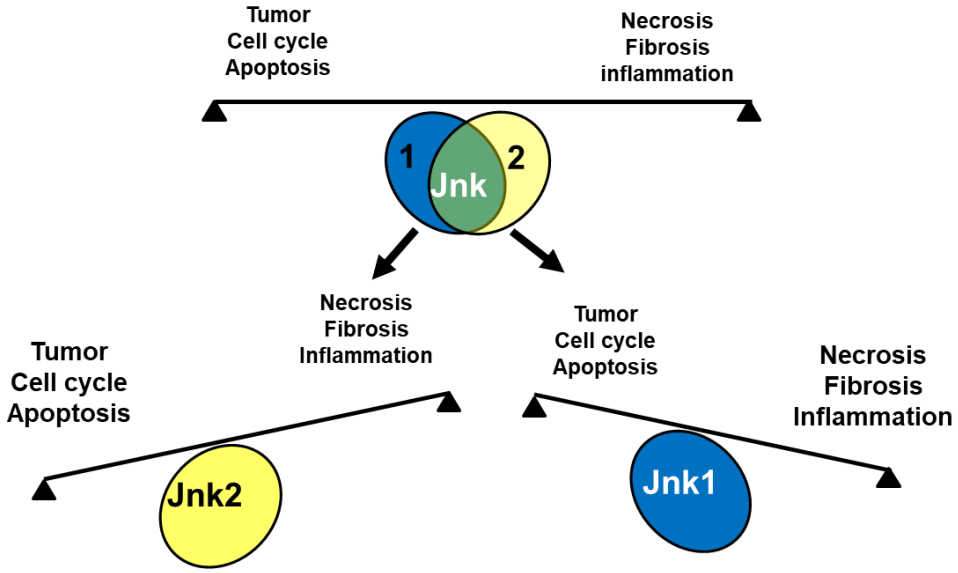
Our first observation with the NEMO mice: A model of hepatocarcinogenesis (HCC)

	8 weeks	26 weeks	52 weeks
NEMO$^{f/f}$			
NEMO$^{\Delta hepa}$			
	early injury cell death	chronic inflammation fibrogenesis	HCC

Differential effects of Jnk1 and Jnk2 on the progression of liver disease







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Epigenetic alterations in CCA: DNA Hypermethylation *DNMTs*, *G9a* and *UHRF1*

Direct interaction between DNMT1 and G9a coordinates DNA and histone methylation during replication.

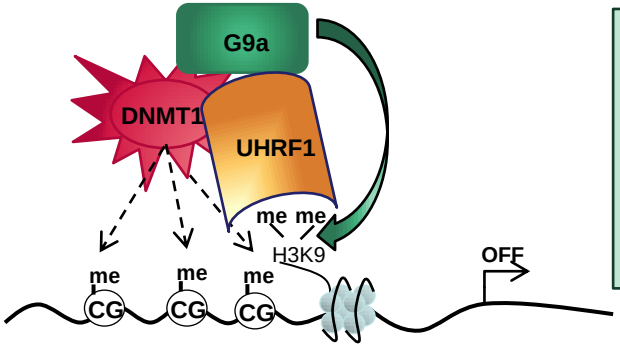
Estève et al, *Genes Dev.* 2006

Aberrant DNA methylation profile in cholangiocarcinoma.

Huang L. et al, *World J Gastrointest Pathophysiol*. 2010

Global Alterations of DNA Methylation in Cholangiocarcinoma Target the Wnt Signaling Pathway.

Goeppert B. et al, *Hepatology, 2013*



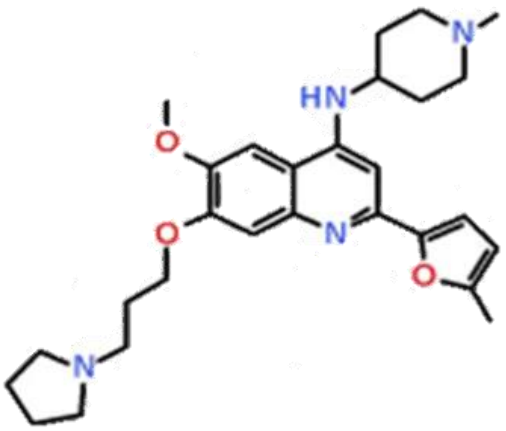
The histone methyltransferase G9a: a new therapeutic target in biliary tract cancer.

Mayr C. et al, Hum Pathol, 2018

CM-272, a dual reversible inhibitor of G9a and DNMT1

- First-in-class **dual** and ***reversible*** inhibitors targeting **G9a** & **DNMTs**.
- Substrate competitive (H3 and DNA).
- Not S-adenosylmethionine (SAMe) competitive.
- Over 200 compounds synthesized/screened for optimal G9a and DNMT inhibition.

San Jose-Eneriz E. *et al*. *Nat. Commun., 2017*



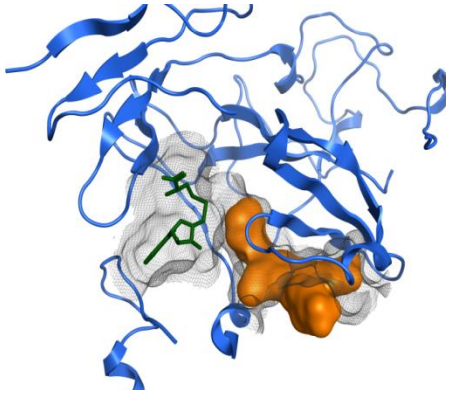
4-aminoquinolines

Lead compound **CM-272**

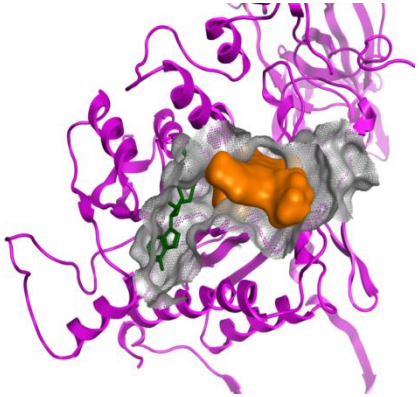
G9a, IC_{50} (nM): 8

DNMT1, IC_{50} (nM): 382

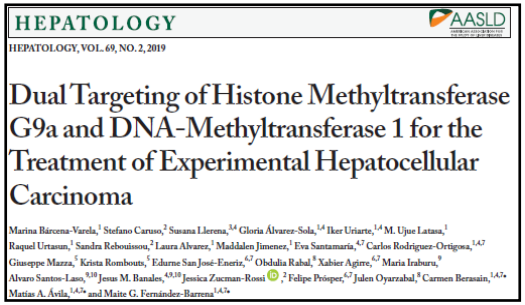
G9a



DNMT1



Plausible binding modes, proposed by docking studies, using known crystal structures (3RJW.pdb for G9a and 4DA4.pdb for DNMT1) for CM-272



HEPATOLOGY

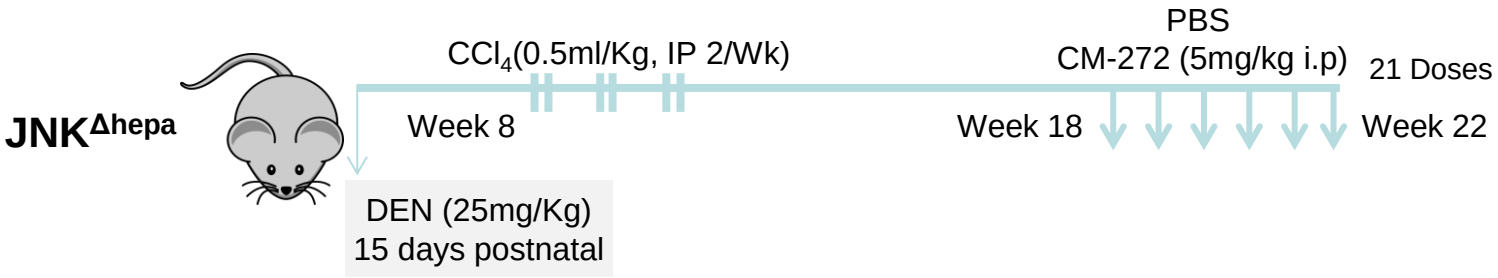
AASLD

HEPATOLOGY, VOL. 69, NO. 2, 2019

Dual Targeting of Histone Methyltransferase G9a and DNA-Methyltransferase 1 for the Treatment of Experimental Hepatocellular Carcinoma

Marina Bárcena-Varela,[1] Stefano Caruso,[2] Susana Llerena,[3,4] Gloria Álvarez-Sola,[1,4] Iker Uriarte,[1,4] M. Ujue Latasa,[1] Raquel Urtasun,[1] Sandra Rebouissou,[2] Laura Alvarez,[1] Maddalen Jimenez,[1] Eva Santamaría,[4,7] Carlos Rodriguez-Ortigosa,[1,4,7] Giuseppe Mazza,[5] Krista Rombouts,[5] Edurne San José-Eneriz,[4,7] Obdulia Rabal,[8] Xabier Agirre,[4,7] Maria Iraburu,[9] Alvaro Santos-Laso,[9,10] Jesus M. Banales,[4,9,10] Jessica Zucman-Rossi,[2] Felipe Prósper,[4,7] Julen Oyarzabal,[8] Carmen Berasain,[1,4,7]* Matías A. Ávila,[1,4,7]* and Maite G. Fernández-Barrena[1,4,7]*

CM-272 inhibits CCA growth in vivo



JNK$^{\Delta hepa}$ mice treated with **DEN/CCl$_4$** develop **intrahepatic cholangiocarcinoma**	Treatment with **CM-272** for 3 weeks ameliorated intrahepatic cholangiocarcinoma formation *in vivo*