ROLE OF IL28B AND ITPA POLYMORPHISMS IN DIFFERENT GENOTYPES OF HCV

(Especially HCV-6)

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HCV GENOTYPES: HK SCENARIO


HCV: HK SCENARIO (QMH)

Genotype 1
(n=220)

Route of Transmission

Genotype 6
(n=101)

(n=78)

- Transfusion: 71%
- IV drug use: 9%
- Others: 20%

p<0.001

Seto WK et al. J Hepatol 2010

- Transfusion: 56%
- IV drug use: 28%
- Others: 16%
HCV-6: MOST COMMON GENOTYPE IN GUANGDONG PROVINCE, CHINA

Prevalence in HCV RNA +ve blood donors (n=270)

Mean age: 34.4 years

Fu et al J Viral Hepat 2011
PHYLOGENETIC ANALYSIS OF HCV-6 IN DRUG USERS (N=210)

Fu et al PLoS One 2012
HCV GENOTYPE 6 – INCREASING PREVALENCE IN SOUTHERN CHINA

Fu et al PLoS One 2012
HCV-1 AND HCV-6: SIMILAR NATURAL HISTORY

Seto WK et al J Hepatol 2010

Probability of cirrhotic complications

Genotype

1  p=0.358

6

Time to develop complications (Years)

Seto WK et al J Hepatol 2010
HCV-1 AND HCV-6: SIMILAR NATURAL HISTORY

Seto WK et al J Hepatol 2010
Future Trends in HCV Therapy

Cure rate


IFN-α2b 24 weeks
IFN-α2b 48 weeks
IFN/RBV 48 weeks
PEG/RBV 48 weeks
Triple Rx
Protease inhibitor + PEG/RBV
24 weeks
PEG/RBV 27%
Triple Rx 75%
NO IFN 12 wks 95-100%
Combo DAA 1st DAA + 2nd DAA
SVR FOR DIFFERENT GENOTYPES
(PEG-IFN AND RIBAVIRIN)

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<thead>
<tr>
<th>Genotype</th>
<th>24 weeks</th>
<th>48 weeks</th>
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<tr>
<td>1</td>
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<td>6</td>
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## SVR for HCV Genotype 6

<table>
<thead>
<tr>
<th>REGION</th>
<th>STUDY</th>
<th>SVR 48 weeks</th>
<th>SVR 24 weeks</th>
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<tbody>
<tr>
<td>Hong Kong</td>
<td>Fung et al J Infect Dis 2008</td>
<td>86%</td>
<td>-</td>
</tr>
<tr>
<td>California, USA</td>
<td>Lam et al Hepatology 2010</td>
<td>79%</td>
<td>70%</td>
</tr>
<tr>
<td>Vietnam</td>
<td>Thu Thuy et al J Hepatol 2012</td>
<td>86%</td>
<td>81%</td>
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Genome-wide association studies (GWAS) involve the genotyping of large case-control cohorts using array-based genotyping platforms typically including several hundred thousand markers. The application of GWAS to a large number of different phenotypes of interest (often specific diseases) in recent years has resulted in the identification of an enormous number of associated loci, which have dramatically advanced understanding of genetic factors contributing to human disease. In this review, the genetic architecture of human disease is addressed and the technical features of GWAS are described. Next, approaches for defining the biologic basis for GWAS signals are described. The future challenges of developing improved phenotypic definitions for complex traits are also discussed.

The genetic architecture of human disease

Most human diseases have a genetic component to their pathogenesis, even infectious diseases typically affected individuals (genetic linkage) narrows the genomic regions in which the disease mutation is likely to reside. Once the likely genomic location has been narrowed, identifying the causative mutation for Mendelian disorders has been, in many cases, relatively straightforward, with a preponderance of missense mutations identified that alter the protein-coding sequence of a gene. The identification of missense mutations within the gene causing cystic fibrosis in 1989 (cystic fibrosis transmembrane regulator [CFTR]) was rapidly followed, in subsequent years, with the identification of many disease-causing mutations for Mendelian disorders. Importantly, in the intervening years, as mapping and sequencing of the human genome advanced, the tools to successfully identify mutations have markedly accelerated the capacity to identify disease-associated mutations for Mendelian disorders.

Complex, multigenic diseases

However, the vast burden of human disease with...
Genome-wide association of IL28B with response to pegylated interferon-α and ribavirin therapy for chronic hepatitis C

Yasuhiro Tanaka,1,18 Nao Nishida,2,18 Masaya Sugiyama,1 Masayuki Kurosaki,3 Kentaro Matsurra,1 Nasuy Sakamoto,1 Mina Nakagawa,4 Masaki Kurenaga,5 Keisuke Hino,5 Shuhei Higé,6 Yoshito Ito,7 Eiji Mita,8 Eiji Tanaka,9 Satoshi Mochida,10 Yoshikazu Murawaki,11 Maqao Honda,12 Akito Sakai,12 Yoichi Hayas,13 Shuhei Nishiguchi,14 Asako Koike,15 Iaso Sakaida,16 Masatoshi Imamura,17 Kiyoseki Ito,17 Koji Yano,17 Naohiko Masaki,17 Fumina Sugauchi,3 Namiki Izumi,3 Katsushi Tokunaga12 & Masashi Mizokami12

The recommended treatment for patients with chronic hepatitis C, pegylated interferon-α (PEG-IFN-α) plus ribavirin (RBV), does not provide sustained virologic response (SVR) in all patients. We report a genome-wide association study (GWAS) to null virologic response (NVR) in the treatment of patients with hepatitis C virus (HCV) genotype 1 within a Japanese population. We found two SNPs near the gene IL28B on chromosome 19 to be strongly associated with NVR (rs12979860, 3.11×10^{-15} and rs8099917, 3.11×10^{-15}). We replicated these associations in an independent cohort (combined P values, 2.84×10^{-27} (OR = 17.7; 95% CI = 10.0–31.3) and 2.68×10^{-32} (OR = 27.1; 95% CI = 14.6–50.3), respectively). Compared to NVR, these SNPs were also associated with SVR (rs12979860, 3.11×10^{-27}; rs8099917, 1.11×10^{-27}). In further fine mapping of the region, seven SNPs (rs8105790, rs11881222, rs8103142, rs28416813, rs4803219, rs8099917 and rs7248668) located in the IL28B region showed the most significant associations (P = 5.52×10^{-26}, 2.66×10^{-32}; OR = 22.3–27.1). Real-time quantitative PCR assays in peripheral blood mononuclear cells showed lower IL28B expression levels in individuals carrying the minor alleles (P = 0.013).

estimated that in 1999, there were 170 million HCV carriers worldwide, with 3–4 million new cases appearing each year. HCV infection affects more than 4 million people in the United States, where it represents the leading cause of cirrhosis and hepatocellular carcinoma as well as the leading cause of liver transplantation. The American Gastroenterological Association estimated that drugs are the largest direct costs of hepatitis C.

The most effective current standard of care in patients with chronic hepatitis C, a combination of PEG-IFN-α with ribavirin, does not produce SVR in all patients treated. Large-scale studies on 48-week-long PEG-IFN-α/RBV treatment in the United States and Europe showed that 42–52% of patients with HCV genotype 1 achieved SVR, and similar results were found in Japan. However, older patients (greater than 50 years of age) had a significantly lower rate of SVR due to poor adherence resulting from adverse events and laboratory-detectable abnormalities such as neutropenia and thrombocytopenia. Specifically, various well-described side effects (such as a flu-like syndrome, hematologic abnormalities and adverse neuropsychiatric events) often necessitate dose reduction, and 10–14% of patients require premature withdrawal from interferon-based therapy. To avoid these side effects in patients who will not be helped by the treatment, as well as to reduce the substantial cost of PEG-IFN-α/RBV treatment, it would be useful to be
IFN-lambda (λ) is a compelling biological candidate

- Type 3 IFN
- IFNλ-1/2/3 = IL29, IL28A, IL28B
- All signal via the IFNL-R
- Expression of IFNL-R restricted
- IFNλ has antiviral activity against HCV

Kelly et al Gut 2011
C allele (rs12979860) is associated with SVR

IL28B-TYPE PREDICTS SVR

- Caucasians: N=1171
  - TT: 27
  - CT: 33
  - CC: 69
  - SVR rate: 33%
  - p<0.0001

- African Americans: N=300
  - TT: 13
  - CT: 15
  - CC: 48
  - SVR rate: 15%
  - p=0.2

- Hispanics: N=116
  - TT: 27
  - CT: 38
  - CC: 56
  - SVR rate: 27%
  - p=0.02

Thompson et al Gastroenterology 2010
**IL28B Polymorphism is Strongest Baseline Predictor of SVR Using PegIFN/RBV**

Odds ratio (95% CI)

- Fasting serum glucose <5.6 mmol/L: $P < 0.0001$
- Hispanic vs. Black: $P = 0.004$
- Metavir F0/F1: $P < 0.0001$
- Caucasian vs. Black: $P < 0.0001$
- VL < 600,000 IU/mL: $P < 0.0001$
- IL28B: CC vs. non-CC: $P < 0.0001$

**Covariates:**
- rs12979860 (2-level), ethnicity (4-level), age (≤ 40), gender, BMI (< 30), VL (≤ 600,000), ALT (≤ ULN), fasting glucose (< 5.6), hepatic steatosis (N/Y [> 0%]), fibrosis (METAVIR F012), RBV (> 13 mg/kg/d)

N = 1,604

The global prevalence of C/T alleles at SNP rs12979860 may explain the recognized geographical variation in SVR rates.
SUMMARY: *IL28B*-TYPE AND GENOTYPE 1 HCV

- is strongly associated with increased SVR rate
- explains much of the ethnic differences in response rates
- is the strongest pre-treatment predictor of SVR
- increases SVR rate 2-fold in non-RVR patients
IL28B predicts SVR?

Yes

Selected Populations

No

Thompson et al Gastroenterology 2010

An IL28B Polymorphism Determines Treatment Response of Hepatitis C Virus Genotype 2 or 3 Patients Who Do Not Achieve a Rapid Virologic Response

Mangia et al Gastroenterology 2010

IL28B Genetic Variation and Treatment Response in Patients with Hepatitis C Virus Genotype 3 Infection

Moghaddam et al Hepatology 2011

IL28B polymorphism is associated with treatment response in patients with genotype 4 chronic hepatitis C

Asselah et al J Hepatol 2012
IL28B genotype is associated with increased SVR in non-RVR G2/3 patients.

Mangia et al. Gastro, 2010
INOSINE TRIPHOSPHATASE (ITPA)

**ITPA gene variants protect against anaemia in patients treated for chronic hepatitis C**

Jacques Fellay¹,* Alexander J. Thompson⁴,* Dongliang Ge¹,* Curtis E. Gumbs¹, Thomas J. Urban¹, Kevin V. Shianna¹, Latasha D. Little¹, Ping Qiu³, Arthur H. Bertelsen⁵, Mark Watson³, Amelia Warner³, Andrew J. Muir⁶, Clifford Brass⁷, Janice Albrecht⁸, Mark Sulkowski⁹, John G. Mc Hutchison² & David B. Goldstein¹

Chronic infection with the hepatitis C virus (HCV) affects 170 million people worldwide and is an important cause of liver-related morbidity and mortality¹. The standard of care therapy combines pegylated interferon (pegIFN) alpha and ribavirin (RBV), and is associated with a range of treatment-limiting adverse effects². One of the most important of these is RBV-induced haemolytic anaemia, which affects most patients and is severe enough to require dose modification in up to 15% of patients. Here we show that genetic variants leading to inosine triphosphatase deficiency, a condition not thought to be clinically important, protect against haemolytic anaemia in hepatitis C-infected patients receiving RBV.

The SNPs showing a genome-wide significant association with quantitative week-4 Hb reduction were spread over a 250 kilobase (kb) region that contains five different protein-coding genes (Fig. 1). We tested the independence of the top association signals in the European-American population using nested linear regression models, in which individual SNPs were added after inclusion of rs6051702, and found evidence for multiple independent signals of association: the most strongly associated SNPs after accounting for the contribution of rs6051702 have P values of 1.4 × 10⁻⁷⁰ (rs2295547) and 2.3 × 10⁻⁴⁷ (rs6051855). The persistence of strongly significant association in the region after accounting for the discovery variant suggests the possibility of multiple causal variants and/or
ITPA VARIANTS PROTECT AGAINST RIBAVIRIN-RELATED ANEMIA IN HCV GENOTYPE 1

Thompson et al Gastroenterology 2010
ITPA RS1127354 possibly predicts SVR in HCV-1

Kurosaki et al, Antivir Ther 2011
Genome-wide association study of interferon-related cytopenia in chronic hepatitis C patients

ITPA Polymorphisms

Platelet

Hb

Thompson et al J Hepatol 2012
HOW ABOUT HCV GENOTYPE 6?

Experience in QMH and PMH
The Effects of IL-28B and ITPA Polymorphisms on Treatment of Hepatitis C Virus Genotype 6

Wai-Kay Seto, MRCP¹, Yasuhiro Tanaka, MD², Kevin Liu, MRCP¹, Ching-Lung Lai, MD¹ and Man-Fung Yuen, MD, PhD¹

doi: 10.1038/ajg.2011.40

Seto WK et al. Am J Gastroenterol 2011
228 patients treated with pegylated interferon and ribavirin

Genotype 1: 105
Genotype 6: 97
Other genotypes: 26

60 patients included in current study

28 patients consent not obtained
6 patients defaulted follow-up
2 patients co-infected with HBV
1 patient co-infected with HIV

Seto et al J Viral Hepat in press
**IL28B**

rs8099917

Chromosome 19

**ITPA**

rs1127354

Chromosome 20

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**Applied Biosystems TaqMan SNP Genotyping Assay**

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Seto et al J Viral Hepat in press
Effect of IL28B and ITPA genotypes on SVR rates

For IL28B Genotype:
- TT (n=52): 100.0% SVR
- TG (n=8): 25.0% SVR

For ITPA Genotype:
- CC (n=37): 100.0% SVR
- CA (n=23): 0.0% SVR

p values:
- IL28B Genotype: p=0.014
- ITPA Genotype: p=0.640
### Factors Associated with SVR

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<thead>
<tr>
<th>Factor</th>
<th>p</th>
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<td>Age</td>
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</tr>
<tr>
<td>Gender</td>
<td>0.601</td>
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<tr>
<td>Type of IFN</td>
<td>0.553</td>
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<tr>
<td>Albumin</td>
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<tr>
<td>Bilirubin</td>
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<tr>
<td>ALT</td>
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<tr>
<td>AST</td>
<td>0.500</td>
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<tr>
<td>Platelet</td>
<td>0.161</td>
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<table>
<thead>
<tr>
<th>Factor</th>
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<tbody>
<tr>
<td>HCV RNA</td>
<td>0.968</td>
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<tr>
<td>APRI</td>
<td>0.617</td>
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<tr>
<td>Ribavirin dose reduction</td>
<td>0.160</td>
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<tr>
<td><em>IL28B</em></td>
<td><strong>0.014</strong></td>
</tr>
<tr>
<td>ITPA</td>
<td>0.387</td>
</tr>
</tbody>
</table>

Seto et al. J Viral Hepat in press
19 patients require reduction of ribavirin dose after median treatment duration of 8 (range 2-32) weeks.

Seto et al J Viral Hepat in press
ITPA genotypes and the median reduction in hemoglobin from baseline

Error bars depict interquartile range

Seto et al J Viral Hepat in press
ITPA genotypes and the median reduction in platelet count from baseline.

- CC genotype (n=37)
- CA genotype (n=23)

Error bars depict interquartile range.

Seto et al J Viral Hepat in press.
THE CONFUSING FUTURE OF HCV THERAPY

Preclinical

Phase I

Phase II

Phase III

Approved

DAA combinations

Nuc-Polymerase inhibitors

Non Nuc-Polymerase inhibitors

Protease inhibitors

Others

DEB025 cyclophilins
IL28B STILL USEFUL IN TRIPLE THERAPY FOR HCV-1 (SPRINT-2 TRIAL)

Poordad et al Gastroenterology 2012

*~90% eligible for short duration therapy
Error bars are 1\(^{\text{st}}\) standard error of mean

**IL28B AND IFN-FREE REGIMENS (DANOPREVIR AND MERICITABINE)**

Chu et al Gastroenterology 2012
CONCLUSIONS

- Prevalence of HCV-6 increasing in Southern China
- IL28B polymorphisms predict SVR in chronic HCV-6
- ITPA polymorphisms predict degree of ribavirin-related anemia in HCV-6
- Usefulness of IL28B in new HCV therapies still present but attenuated
ACKNOWLEDGEMENTS

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- Prof. CL Lai
- Dr. Kevin Liu
- Dr. Owen TY Tsang (PMH)
- Dr. Jacky MC Chan (PMH)
THANK YOU!