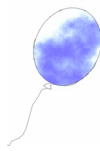


HEPATITIS C CARING AMBASSADORS PROGRAM NEWSLETTER

April 2004

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IN THE NEWS

Hepatitis C scare halts use of surgical adhesive

<http://www.yomiuri.co.jp/newse/20050427wo71.htm>

[Japan] The Health, Labor and Welfare Ministry on Tuesday ordered a pharmaceutical company to stop the sale of an imported surgical tissue adhesive after an elderly man was found to have contracted the hepatitis C virus after his liver functions weakened following an operation. The man, who is in his 70s, suffered decreased liver function following an operation in May last year, during which the tissue adhesive Takokonbu was used. One of the basic ingredients in Takokonbu is fibrinogen, which is found in human blood, and is culled from blood supplies in the United States. The adhesive is used to prevent blood and other bodily fluids from leaking from internal organs during surgery. Takokonbu has been shipped to medical institutions throughout the country.

Wait twelve weeks before treating acute HCV in HIV-positive patients. Gilleece YC et al. Is the treatment of acute hepatitis C in HIV-positive individuals effective?. Eleventh Annual Conference of the British HIV Association, Abstract O26, Dublin, April 20 – 23rd, 2005.

<http://www.aidsmap.com/en/news/414EA10C-33FB-4689-B586-8DB5F4A2CEF6.asp>

Treatment for primary hepatitis C virus infection is significantly less likely to be effective in coinfecting HIV-positive individuals, than patients who are hepatitis C monoinfected, according to a study presented to the Eleventh Annual Conference of the British HIV Association in Dublin on April 21st. In addition, investigators found that a significant number of HIV-positive patients spontaneously cleared hepatitis C virus within the early weeks of infection, and accordingly they recommend that if treatment for hepatitis C is initiated during acute infection, it should be delayed for twelve weeks.

Approximately 90% - 98% of individuals who are monoinfected with hepatitis C virus achieve a sustained virological response after receiving anti-hepatitis C virus therapy consisting of pegylated interferon during the acute infection period. There are limited data about the effectiveness of this strategy in HIV-positive patients. It is known however that during acute infection with hepatitis C virus, the HIV-positive individuals have significantly higher hepatitis C viral loads than individuals infected only with hepatitis C.

In a prospective, open label study investigators from the Chelsea and Westminster Hospital in London evaluated the efficacy and safety of treatment for hepatitis C virus during acute infection. Between 1997 and 2003, a total of 50 gay men with a mean age of 37 years were recruited for the study. Hepatitis C virus infection was confirmed by antibody testing. Hepatitis C virus viral load was measured at baseline, and at weeks four, twelve, 24, 32 and 48.

If an individual was still hepatitis C virus RNA-positive at week 12 they were treated with pegylated interferon and ribavirin for 24 weeks. The primary endpoint of the study was the proportion of patients with a sustained virological response to treatment (defined as a negative hepatitis C virus PCR test at 48 weeks). Data on CD4 cell count, HIV viral load, liver function, side-effects and laboratory abnormalities were also gathered. Of the 50 men included in the investigators' analysis, 12 (24%) spontaneously cleared hepatitis C virus infection by week twelve and therefore did not receive anti-hepatitis C therapy. Spontaneous clearance of hepatitis C virus was significantly associated with a CD4 cell count above 500 cells/mm³ (p = 0.03), and a lower hepatitis C viral load (p = 0.04).

A total of 27 men accepted anti-hepatitis C therapy, 16 (59%) of whom achieved a sustained virological response, and this was significantly associated with a lower mean peak ALT level (p < 0.001). Due to the small number of patients in the study, it was not possible to say if treatment outcome was significantly different between individuals infected with different hepatitis C virus genotypes. Median CD4 cell count fell by 58 cells/mm³ on the completion

of anti-hepatitis C therapy, and the level of HIV viral load did not change significantly. These are usual accounts for individuals receiving hepatitis C virus treatment. No deaths or new opportunistic infections occurred. Side-effects were widely reported, the most common being depression and anxiety (33 instances) and flu-like symptoms (22 individuals). In addition, 13 cases of neutropenia and three cases of anaemia were recorded. One patient had to discontinue his treatment because of side-effects.

The investigators concluded that as only 59% of HIV-positive patients who are treated for acute hepatitis C virus infection achieve a sustained virological response, this approach to the management of hepatitis C is less effective in coinfecting patients than hepatitis C virus mono-infected individuals.

Hepatitis C Can Be Sexually Transmitted

<http://www.reuters.com/newsArticle.jhtml?type=healthNews&storyID=8287010>

The virus that causes hepatitis C (HCV) can be sexually transmitted and the risk is directly related to the frequency of intercourse, the findings from a new study suggest. However, the association does not seem to be very strong. Contact with infected blood is the main route by which HCV is transmitted, lead author Dr. Veysel Tahan, from Marmara University in Istanbul, Turkey, and colleagues note. However, in up to 50 percent of cases, a transmission route is never identified.

Researchers have hypothesized that some of these unknown cases may be caused by sexual transmission. To investigate this controversial topic, Tahan's team assessed HCV transmission between 600 patients with chronic hepatitis C and their spouses. In a subgroup of 216 spouses who were HCV-negative at the start of the study, the authors assessed transmission rates over an average period of 36 months. The findings appear in the current issue of the American Journal of Gastroenterology.

Antibodies against HCV, indicating exposure to the virus, were detected in two percent of the spouses, the researchers note. The average age and duration of marriage were not associated with the likelihood of detecting HCV antibodies. In contrast, an association between the frequency of sexual intercourse and HCV transmission risk was observed. However, this relationship was not statistically significant, indicating the association was. In the subgroup of 216 spouses, none of the HCV-negative subjects became positive during follow-up, the investigators note.

"The risk of HCV transmission between monogamous sex partners may be related to the number of sexual intercourse" events, the authors state. "The absence of any evidence of transmission in the three-year follow-up period suggests that this relation is not very strong." In a related editorial, Dr. Norah A. Terrault, from the University of California at San Francisco, comments that the "low incidence of new infections among monogamous couples supports the clinical practice of not recommending that such couples change their sexual practices."

Aethlon Medical Sends HIV-AIDS & Hepatitis-C Treatment Technology to India

http://home.businesswire.com/portal/site/google/index.jsp?ndmViewId=news_view&newsId=20050421005534&newsLang=en

April 21, 2005--Aethlon Medical, Inc. announced that it will begin shipping its Hemopurifier(TM) treatment technology to India today. The Company stated that today's shipment is the first lot of Hemopurifiers that will be sent to support the HIV and Hepatitis-C human studies that were announced in February. Aethlon Chairman & CEO, James A. Joyce stated, "The initial shipment of our Hemopurifier treatment technology is a significant milestone for our research team, especially when considering that each Hemopurifier represents new hope for infected individuals that don't have access or are unresponsive to the current standard of care." Once received in India, the initial Hemopurifier lot will undergo required biocompatibility and toxicity testing prior to initiating human studies. Aethlon Chief Scientific Officer, Dr. Richard Tullis added, "Our manufacturing and quality procedures are in place and operating efficiently. We are on track to achieve safety and preliminary efficacy data this summer."

Grits agree to extend hepatitis C benefits

<http://www.theglobeandmail.com/servlet/story/RTGAM.20050420.wxhepc0421/BNStory/National/>

[Canada] The federal Liberals have reversed an eight-year stand against extending support to the "forgotten victims" of Hepatitis C, a move the opposition attributed to election fears rather than compassion. All parties voted in favour yesterday of a motion brought by Stephen Fletcher, the Conservative health critic, to give financial help to people who contracted the disease through tainted blood before 1986 or after 1990. An earlier \$1.1-billion package had compensated only those who were infected between those years. Mr. Fletcher charged that the Liberals

reversed their policy because refusing to do so would hurt them in an election anticipated in June. "Let's face it, it would have been hugely embarrassing for the Liberals to vote against this motion," Mr. Fletcher told a news conference. "They'll do anything to try to keep power."

The motion says that given a large surplus in the fund established to compensate the original group of victims — an amount pegged at about \$865 million — the fund should be used immediately to compensate those left out of the agreement. The motion was to have been voted upon in the Commons two weeks ago, but only three hours of debate are allowed on such items. When the House broke for the day, there were still two minutes to go. The Liberal government allowed no extension. Yesterday's vote ended years of Liberal resistance to compensating all victims of hepatitis C who contracted the disease through tainted blood. But the apparent policy reversal does not mean people who contracted the disease before 1986 or after 1990 will be paid soon. Health Minister Ujjal Dosanjh said in November that he was prepared to look at the options for helping those left out of the original agreement. He said yesterday that the Liberal government agreed to vote for Mr. Fletcher's motion, despite it being badly flawed. There will have to be lengthy discussions to determine who qualifies under this new category of hepatitis C victim, said Mr. Dosanjh, so it cannot be paid out immediately. And "the motion talks about in recognition of a surplus. We don't know whether there's a surplus. We will only know whether there's a surplus when the court determines there's a surplus." The lawyers who settled the class-action lawsuits for those victims who contracted the disease from 1986 to 1990 are vigorously opposed to extending the compensation to others. The government is in negotiation with those lawyers to determine what is left in the fund, Mr. Dosanjh said. But he promised that all those who contracted the disease through tainted blood eventually would get compensation. "We want to make sure that this is done," Mr. Dosanjh said.

Patients alerted over Hepatitis C: A group of women who were treated at three Scottish hospitals are being screened for Hepatitis C.

http://news.bbc.co.uk/2/hi/uk_news/scotland/4460381.stm

The 67 women are among 2,000 in the UK who were patients of a gynaecologist who had the virus. The doctor worked at 25 hospitals in England and three in Scotland - Forth Park and the Victoria in Kirkcaldy and the Royal Alexandra in Paisley. Health officials insisted the risk of infection was very small and screening was being undertaken as a precaution.

Officials from Health Protection Scotland have contacted 41 patients treated by NHS Fife in 1986 and 26 patients who were treated by NHS Argyll and Clyde in 1997 and 1998. They have received letters, warning them they could be at risk. The women have been asked to take a blood test to make sure they are safe and have been offered counselling.

The gynaecologist was immediately transferred to a healthcare area where patients were not at risk. NHS Argyll & Clyde's consultant in public health medicine, Catherine Chiang, said: "Like most people who are infected with hepatitis C, the healthcare worker had no symptoms and was unaware of the infection." Dr. Lesley MacDonald, NHS Fife's director of public health, said: "We have worked hard to carefully identify any patient who might have been at risk of contracting infection with hepatitis C."

Human Genome Sciences Hepatitis C Drug Passes Test

<http://www.reuters.com/newsArticle.jhtml?type=healthNews&storyID=8185669>

Human Genome Sciences Inc. on Thursday said its experimental drug for hepatitis C met its primary goal in a mid-stage trial of patients who had not previously been treated for the liver-damaging virus. The Rockville, Maryland-based company said the favorable results were seen in a trial of 56 patients with the genotype 1 strain of hepatitis C, the hardest to treat of three main strains, who were given varying doses of its Albuferon injectable drug. The patients had never previously been treated for their condition.

Albuferon is a laboratory-altered form of interferon-alpha, a standard treatment for hepatitis C. But unlike standard interferon treatments that must be injected once weekly, researchers said the Phase II trial indicated Albuferon is highly effective with injections only every two to four weeks. Patients taking the two highest doses of the drug saw their levels of virus decline by over 99.9 percent 28 days after treatment began. That satisfied the trial's primary goal of showing at least a 99 percent reduction in virus, meaning a reduction to undetectable levels, after four weeks.

The company said 69 percent of patients in those two high-dose groups achieved the 99 percent reduction of virus after four weeks. Undetectable virus levels after 42 days were seen in 23 percent of the same patients. Swiss drugmaker Roche sells the most popular interferon, called Pegasys, which is taken with a pill called ribavirin to

treat the often-deadly virus. Schering-Plough Corp.'s interferon, called Peg-Intron, is also taken alongside ribavirin. Those standard combo treatments are considered the best available therapies for the virus, but use of them for up to 48 weeks only knocks down the virus to undetectable levels in about 42 percent of patients with genotype 1, Human Genome Sciences said.

New Data on Most Prescribed Hepatitis C Therapy Pegasys(R) Presented at European Association for the Study of the Liver (EASL) Conference: Roche continues pioneering research to enhance treatment for hepatitis C patients

<http://biz.yahoo.com/prnews/050415/nyf011.html?.v=5>

New Pegasys® (peginterferon alfa-2a) data, including exploration of factors associated with treatment response in hepatitis C patients, were revealed in 33 oral and poster presentations at the 40th Annual Meeting of The European Association for the Study of the Liver, April 13 - 17, in Paris, France.

Data from several Pegasys studies will be presented and discussed at the meeting, including:

1. *The Influence of Cumulative Peginterferon Alfa-2a (40 KD) and Ribavirin (RBV) Exposure on Sustained Virological Response (SVR) Rates in Patients with Genotype 1 Chronic Hepatitis C* This analysis of pooled data from two phase III trials looked at the relationship between cumulative ribavirin exposure (total drug administered) and the desired outcome (sustained virological response, or SVR) in 569 chronic hepatitis C patients treated with Pegasys and ribavirin. The analysis found that as the cumulative dose of ribavirin decreased, SVR rates also decreased. Sixty-six percent of patients who took greater than 97 percent of the recommended ribavirin dose achieved an SVR compared to only 33 percent in patients who took less than 60 percent of the recommended ribavirin dose.

2. *Age and Sustained Virological Response in Patients With Persistently 'Normal' ALT and Chronic Hepatitis C Treated With Peginterferon Alfa-2a (40KD) Plus Ribavirin* A retrospective analysis of 422 chronic hepatitis C patients with persistently 'normal' ALT levels showed that younger patients had higher response rates to Pegasys and Copegus treatment compared to older patients. In the study, patients were divided into two groups based on age (<40 or >40), and were treated for 24 or 48 weeks. The study found that 54 percent of patients in the younger group achieved an SVR compared to 34 percent of those in the older group.

3. *Clustering of Poor Prognostic Factors in Patients with Chronic Hepatitis C* An analysis of 2,404 patients from two multinational, randomized, controlled phase III studies showed that a cluster of poor prognostic characteristics, and not weight alone, may explain why lower response rates to therapy are observed in heavier patients, irrespective of the pegylated interferon therapy used. Patients were divided into three weight categories: <65 kg, <65-<85 kg, >85 kg. As previous studies with both pegylated interferons have shown, the patients in the highest weight category experienced lower SVR rates than lighter weight patients. The heaviest patients were also more likely to be male, black, cirrhotic, and infected through intravenous drug use, which are characteristics that are typically associated with lower treatment response rates.

McGuinty government appoints members of Hepatitis C Task Force: Chair, 16-Member Panel To Examine Hepatitis C Prevention And Treatment [Canada]

<http://www.newswire.ca/en/releases/archive/April2005/04/c1850.html>

The McGuinty government is appointing 16 members to the Hepatitis C Task Force to advise the government on how to improve hepatitis C prevention and treatment, Health and Long-Term Care Minister George Smitherman announced today. "I am delighted that the task force, chaired by John Plater, President of Hemophilia Ontario, will get down to providing advice to the government on strategies to improve hepatitis C treatment, prevention, education and support in Ontario," said Smitherman.

Appointments were effective April 1, 2005. All members have been asked to serve a minimum two-year term, with a maximum term lasting three years, at which time membership of the task force will be reviewed. "The minister has appointed an excellent group of people to the task force; they represent the diversity of expertise and understanding it will take to respond to hepatitis C," said Plater. "The groundwork we lay starting today will not only be of benefit to people living with hepatitis C, but will also reduce the number of Ontarians contracting hepatitis C in the future." Today's announcement fulfils the commitments made by Smitherman last October as part of an action plan to improve hepatitis C care and prevention. The action plan includes:

- Establishing a Hepatitis C Task Force chaired by John Plater to advise the government on strategies to improve the prevention and treatment of hepatitis C

- Creating a Hepatitis C Secretariat within the Ministry of Health and Long-Term Care to support the task force.

Hepatitis C action 'inadequate': The government is not doing enough to stem a "tidal wave" of hepatitis C in the UK, MPs have said.

<http://news.bbc.co.uk/1/hi/health/4369199.stm>

A report from the All-Party Parliamentary Group (APPG) on Hepatology said an action plan launched last year was "inadequate." The committee of MPs called on the government to revise its plans as a "matter of urgency."

Hepatitis C is a blood-borne virus thought to affect around 500,000 people in the UK. But experts warn there are hundreds of thousands of people with the disease who had not yet been diagnosed. The committee said the action plan put together by the Chief Medical Officer for England, Sir Liam Donaldson, set no targets and that the awareness campaign had been very low key. The MPs said there had to be greater urgency in dealing with "the coming tidal wave of hepatitis C" in the UK.

The committee's report - 'The Hepatitis C Scandal' - calls for greater investment from the government to deal with the virus and a proactive screening programme to target at-risk groups. It said these should include women who have had a Caesarean birth and people who received a blood transfusion before screening for the virus was introduced in 1991. David Amess, chairman of the APPG, said: "This report highlights the need for the NHS, and the individual Primary Care Trusts within it, to move hepatitis C much further up the agenda. "It became clear during our investigation that the levels of service available were inadequate to cope well with even the current level of treatment offered to the 10-20% of people living with the virus who have been diagnosed. "The government should review as a matter of urgency both current service provision and plan for the inevitable increased burdens."

Charles Gore, chief executive of leading the Hepatitis C Trust, said: "We are going to have to face the consequences of this disease and it is better to do it sooner rather than later. "There are hundreds of thousands of people out there with hepatitis C who at the moment are undiagnosed and are a risk to themselves because they are not getting treatment, and a risk to others because they could infect them. A Department of Health spokesperson said: "Our Hepatitis C Action Plan will help ensure that people who are infected with the disease are referred for specialist assessment and treatment and also take precautionary measures so that their infection isn't passed on to others. "The awareness-raising campaigns for health professionals and the public are a crucial factor in helping combat the disease." She said the Action Plan which was currently being implemented across the NHS also set out a framework for improvements to prevention, diagnosis and treatment services and to boost research.

Hepatitis C Antibodies May Be Slow to Appear

<http://www.reuters.com/newsArticle.jhtml?type=healthNews&storyID=8027746>

The findings of a long-term study of injection drug users newly infected with hepatitis C virus (HCV) show that antibodies to the virus may not appear in the blood until two months after they have been infected. These results "underscore the importance" of nucleic acid screening of blood donations to prevent HCV transmission, researchers say. The findings also reaffirm the need to follow HCV-positive injection drug users long-term to check for viral persistence.

Among 179 HCV antibody-negative injection drug users followed in the study, an "alarming" 34 percent became infected despite risk reduction counseling, Dr. Andrea L. Cox, from the Johns Hopkins Medical Institutions in Baltimore, Maryland, and colleagues report in the medical journal *Clinical Infectious Diseases*. As expected, individuals with early-phase HCV infection were largely without symptoms. Virus in the bloodstream was the earliest sign of HCV infection, preceding the detection of HCV antibodies by 5 to 6 weeks, and in one case, by more than 12 months, the investigators report. However, in all other cases, HCV RNA was detected no more than 63 days before antibodies appeared.

Liver enzyme function measurements were also elevated during early-phase HCV infection, but did not correlate closely with HCV RNA levels or viral persistence. None of the infected patients developed jaundice. In cases of viral persistence, stable blood levels of HCV RNA were noted in some individuals within 2 months after the virus was first detected in the blood, while in others, it was not apparent until more than 1 year later. In individuals who cleared the virus from their system, HCV became persistently undetectable as early as 94 days and as late as 620 days after initial infection.

Hepatitis C Treatment Makes Economic Sense [New Zealand]

<http://www.scoop.co.nz/stories/GE0504/S00002.htm>

Research by the Christchurch School of Medicine and Health Sciences has shown that methadone maintenance therapy for injecting drug users saves lives, and it is also cost-effective to treat their Hepatitis C virus. The Otago University research team investigated both Maori and non-Maori drug users who are on methadone maintenance and found that excess mortality from drug overdoses was reduced by 75% amongst people who are on methadone maintenance. 98% of the participants stopped injecting drugs, reported large improvements in their health and also reductions in drug-related crime.

Although 84% of the estimated 19,000 injecting drug users in N.Z. have Hepatitis C, few are receiving treatment for the virus. The total number with the virus in NZ is estimated at 30,000 people, but that is expected to double by 2010. End stage Hepatitis C can be fatal, has major effects on the liver and other serious health impacts. There is evidence that large numbers of drug users actually want treatment for Hepatitis C, but don't get it. Meanwhile there are also long waiting lists for methadone maintenance treatment (MMT).

Lead investigator Dr Ian Sheerin from the Department of Public Health and General Practice at the School says there is an urgent need for policies aimed to control the spread of this debilitating virus. These include needle exchanges, education about risky behaviours, blood awareness, access to drug and alcohol services, as well as Hepatitis C treatment. "The size of the health problem is not being matched by government investment in communicable disease control. The policy documents say the right things, but the commitment has not been made to address the issue adequately." "However, the government made an important step in 2004 when it funded combination medication for Hepatitis C. Research shows this treatment is effective. For some of types of Hep C, combination therapy has been shown to cure 80% of cases. "

Dr Sheerin says the key is that although Hepatitis C medication is expensive, the courses are short and will prevent 39% of future health costs. He says if nothing is done, then two things are likely to happen. Hepatitis C will spread, not only amongst drug users, but also in wider N.Z. society. This will have rapidly escalating health and social welfare costs as sufferers become more debilitated, need constant health care, and their livers fail. These costs have already been estimated to rise between to \$166 and \$400 million in the next 30 years unless adequate treatment and disease control is provided. It costs more than \$120,000 for a liver transplant and there is a major shortage of liver donors. It makes more sense to invest in Hepatitis C treatment at an early stage, than wait until people need a liver transplant.

"There's a tendency in N.Z. to see this as just a drug users' problem, therefore why worry. That's ignoring the fact that Hepatitis C is a growing and expensive health issue, which won't go away," says Dr Sheerin. "The existence of a large pool of untreated infectious disease is a potential threat to everyone". Because, the Hepatitis C virus is slow in its progression, there has been limited recognition of this public health issue and New Zealand is currently losing ground by about 1300 new infections every year.

Valeant Pharmaceuticals Presents Final Analyses of Viremagine Phase 2 Clinical Trial at EASL; Viremagine Phase 2 SVR Data Presented in Detail; Four Valeant Abstracts Accepted for Presentation at EASL

<http://www.valeant.com/mediaCenter/newsArticle/newsArticle.jspf?objectId=4096>

Valeant Pharmaceuticals International presented the final analyses of sustained viral response (SVR) and relapse rate data from its Viremagine(R) Phase 2 clinical trial at the 40th Annual Meeting of the European Association for the Study of the Liver (EASL) in Paris, France. The company is developing Viremagine(R), a nucleoside (guanosine) analog, in oral form, currently in Phase 3 clinical trials, for administration in combination with pegylated interferon for the treatment of chronic hepatitis C in treatment-naive patients.

The results of the Phase 2 proof-of-concept study validate the study design by continuing to show that Viremagine demonstrates a significantly reduced incidence of anemia and comparable efficacy to ribavirin. The Viremagine Phase 2 study, conducted entirely in the United States, consisted of 180 treatment-naive subjects with chronic HCV. Treatment duration was based on genotype, with genotypes 2 and 3 receiving 24 weeks of treatment and genotype 1 receiving 48 weeks of treatment, each with a post-treatment follow-up period of 24 weeks. The 24-week follow-up period is considered the standard evaluation of efficacy.

As previously reported, anemia rates (Hb less than 10g/dL) during the treatment period were significantly lower in patients treated with Viremagine than those treated with ribavirin (4 percent versus 27 percent; p less than 0.001). Among patients receiving the 400 mg BID dosage of Viremagine, there were no cases of defined anemia and among patients receiving the 600 mg BID dosage there was only one case of defined anemia (2 percent). In contrast, there was an 11 percent incidence of defined anemia in the 800 mg BID arm and a 27 percent incidence in the ribavirin arm. Other types of adverse events were similar between treatment arms. The most common other adverse events

associated with combination therapy are fatigue, headache, insomnia, depression and myalgia. At the end of the 24-week follow-up period, the SVR rates among the Viramidine arms and ribavirin were not statistically different.

CLINICAL TRIALS, COHORT STUDIES, PILOT STUDIES

Etanercept as an adjuvant to interferon and ribavirin in treatment-naive patients with chronic hepatitis C virus infection: a phase 2 randomized, double-blind, placebo-controlled study. Zein NN, et al. *J Hepatol.* 2005 Mar;42(3):315-22.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15791697

BACKGROUND/AIMS: Current therapies for patients with chronic hepatitis C virus (HCV) do not achieve sustained viral clearance in most patients, and are associated with severe toxic effects. Our aim was to investigate the efficacy and safety of etanercept as adjuvant to interferon and ribavirin in treatment-naive patients with HCV. **METHODS:** Double-blind, randomized, placebo controlled trial. Fifty patients with chronic HCV were randomly assigned to receive interferon alfa-2b and ribavirin with either etanercept or placebo for 24 weeks. The main outcome measure was the absence of HCV RNA at 24 weeks, the on treatment response at the end of the etanercept randomization period. **RESULTS:** At 24 weeks, HCV RNA was absent in 63% (12/19) etanercept patients compared to 32% (8/25) placebo patients ($P=0.04$). In addition, patients receiving etanercept had lower frequency of most adverse events categories compared to placebo. **CONCLUSIONS:** Etanercept given for 24 weeks as adjuvant therapy to interferon and ribavirin significantly improved virologic response at the end of the etanercept randomization period among patients with HCV, and was associated with decreased incidence of most adverse effects associated with interferon and ribavirin.

Is the management of hepatitis C patients appropriate? A population-based study. Hatem C, et al. *Aliment Pharmacol Ther.* 2005 Apr 15;21(8):1007-15.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15813837

[France] **Summary Background :** In order for hepatitis C patients to receive antiviral treatment, they must reach medical care. **Aim :** To assess the proportion of patients reaching medical care after hepatitis C diagnosis in a general population (1 006 171 inhabitants) in France. **Methods :** Between 1994 and 1999, 1508 cases were diagnosed, of which 1251 were eligible for the study. **Results :** Two-hundred and two patients did not have any medical care; among them, 55.4% had normal alanine transferase, 58.4% had risk factors related to lifestyle and 22.8% were alcoholics. Amongst the 1049 other patients, 41.6% had a liver biopsy, 25.0% were treated. Treatment was more often carried out in males than in females (OR: 1.59; $P = 0.001$), and in patients under 65 than in older patients (OR: 2.22; $P < 0.008$). Among non-treatment reasons, alcoholism ($P = 0.001$), drug-addiction ($P = 0.04$) and escaping monitoring ($P = 0.04$) were more frequent in males than in females, whereas normal alanine transferase was more frequent in females than in males ($P = 0.004$). Amongst 278 patients with a Metavir score $>A1F1$, 71 (25.5%) did not undergo treatment. **Conclusion :** In a general population, one patient in six did not receive on-going health care; a quarter of patients with a Metavir score $>A1F1$ did not receive any treatment. These results showed insufficient clinical management, which could compromise the effectiveness of treatment in general population.

Seroprevalence of hepatitis C virus and associated risk behaviours: a population-based study in San Juan, Puerto Rico. Perez CM, et al. *Int J Epidemiol.* 2005 Mar 31; [Epub ahead of print].

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15802378

BACKGROUND: Limited information about the epidemiology of hepatitis C virus (HCV) infection is available in Puerto Rico, one of the areas hardest hit by the human immunodeficiency virus/acquired immune deficiency syndrome (HIV/AIDS) epidemic. We estimated the prevalence of HCV infection and identified correlates of seropositivity in the municipality of San Juan, Puerto Rico. **METHODS:** A probability cluster design was employed to select a sample of households representative of the population aged 21-64 years in San Juan during 2001-2002. All 964 subjects completed a face-to-face interview to gather data on demographics and self-reported risk behaviours followed by venipuncture for HCV antibody testing. Variables that were at least marginally associated with HCV seroprevalence ($P < 0.10$) in the bivariate analyses were considered for inclusion into the multiple logistic regression model to estimate the adjusted prevalence odds ratio (POR). **RESULTS:** Overall weighted prevalence of HCV infection was 6.3% (95% CI 3.6-10.9%). A significant ($P < 0.05$) higher prevalence was observed among subjects with the following characteristics: age 30-49 (9.5%), male sex (10.6%), ≤ 12 years

of education (9.6%), no health coverage (12.6%), lifetime heroin use (39.2%), lifetime cocaine use (39.6%), tattooing practices (34.2%), history of imprisonment (32.8%), and self-reported histories of hepatitis B virus infection (30.4%) and HIV/AIDS (92.1%). Multivariate logistic regression revealed that tattooing practices (POR = 8.9; 95% CI 1.7-44.7), lifetime cocaine use (POR = 5.5; 95% CI 2.2-13.5), blood transfusions prior to 1992 (POR = 4.0; 95% CI 1.6-10.1), lifetime heroin use (POR = 3.3; 95% CI 1.4-7.8), and history of imprisonment (POR = 2.3; 95% CI 1.1-4.9) remained significantly associated with HCV seropositivity. **CONCLUSIONS:** The large prevalence of HCV infection observed in Puerto Rican adults residing in San Juan suggest that HCV infection is an emerging public health concern and merits further investigation.

Usefulness of combined measurement of serum bile acids and ferritin as additional prognostic markers to predict failure to reach sustained response to antiviral treatment in chronic hepatitis C. Jorquera F, et al. *J Gastroenterol Hepatol.* 2005 Apr;20(4):547-54.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15836702

Abstract Aim: To investigate the relationship between serum levels of ferritin and bile acids (BA) and the response to antiviral treatment in chronic hepatitis C (HCV). **Methods:** A retrospective study was carried out on 35 control volunteers and 50 patients receiving interferon alpha-2b alone or plus ribavirin for 48 weeks. These were classified as sustained responders (SR) for >6 months after therapy (n = 17), non-responders (NR) (n = 27) and relapsers (RL) (n = 6). Before treatment, serum ferritin levels were determined by immunoturbidometry, 3alpha-hydroxyl-BA levels (S-3alpha-OH-BA) were assayed enzymatically and total (desulfated, deglucuronidated and deamidated) BA concentrations (STBA) by gas chromatography-mass spectrometry. **Results:** STBA were lower in controls than in patients (SR < NR + RL). The highest levels of cholic acid and chenodeoxycholic acid families were found in NR + RL. Levels of cholic acid family were similar in controls and SR, whereas those of chenodeoxycholic acid family were higher in SR than in controls. A significant correlation between STBA (but not S-3alpha-OH-BA) and ferritin was found. Apparent value to predict the absence of a sustained response was calculated by combining elevated ferritin (>300 microg/mL) and STBA or individual BA species at different cut-off values. The best degree of certainty (100% specificity) was obtained using STBA >15 microM. **Conclusion:** These results recommend that larger prospective trials should be performed in chronic HCV patients to evaluate the usefulness of combined measuring of STBA and ferritin as additional prognostic markers to predict the existence of a very low probability of a sustained response to the current standard treatment, i.e. pegylated interferon in combination with ribavirin.

Pegylated versus standard interferon-alpha in antiviral regimens for post-transplant recurrent hepatitis C: Comparison of tolerability and efficacy. Toniutto P, et al. *J Gastroenterol Hepatol.* 2005 Apr;20(4):577-582.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15836706

Abstract Background: In the treatment of hepatitis C virus (HCV) infection, regimens including pegylated interferon-alpha are superior to those including standard interferon; the present retrospective study was performed to verify whether the same is applicable to biopsy-proven recurrent hepatitis C (genotype 1b) after liver transplantation (OLT). **Methods:** Twenty-four patients (16 male) were studied. Twelve had received interferon-alpha(2b) (IFN), 9 MU weekly and 12 received pegylated interferon-alpha(2b) (PEG-IFN), 0.5 microg/kg weekly. All had received oral ribavirin 600-800 mg/day. Treatment duration was intended for 12 months. A repeat liver biopsy, with evaluation of the Ishak grading and staging scores, was obtained at 1 year. **Results:** Only 12/24 patients (50%) completed a full year of therapy; 17 (71%) experienced side-effects requiring a 50% dosage reduction or discontinuation of the IFN, PEG-IFN and/or ribavirin. This was observed in 6/12 patients (50%) treated with IFN in comparison to 11/12 patients (92%) treated with PEG-IFN (P < 0.05). The difference was mainly accounted for by anemia and leukopenia that were reported in 4/12 IFN patients (33%) versus 9/12 PEG-IFN patients (75%; P < 0.05), respectively. End-of-treatment viral response (ETVR) and histological response were always associated and occurred in 4/24 patients (17%), two in each treatment arm. Patients with ETVR were younger, had always completed 1 year of therapy, had had recurrent hepatitis later after transplantation and presented a higher baseline grading score. **Conclusions:** In the OLT setting, the potential benefits of antiviral treatments including PEG-IFN may be limited by the poor tolerability of the adopted drugs.

The accuracy of the report of hepatic steatosis on ultrasonography in patients infected with hepatitis C in a clinical setting: A retrospective observational study. Hepburn MJ, et al. *BMC Gastroenterol.* 2005 Apr 13;5(1):14 [Epub ahead of print].

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15829009&dopt=Abstract

BACKGROUND: Steatosis is occasionally reported during screening ultrasonography in patients with hepatitis C virus (HCV). We conducted a retrospective observational study to assess the factors associated with steatosis on ultrasonography and the relationship between steatosis on ultrasound versus biopsy in patients infected with HCV in a clinical setting. Our hypothesis was ultrasonography would perform poorly for the detection of steatosis outside of the context of a controlled study, primarily due to false-positive results caused by hepatic fibrosis and inflammation. **METHODS:** A retrospective review of ultrasound reports was conducted on patients infected with HCV in a tertiary care gastroenterology clinic. Reports were reviewed for the specific documentation of the presence of steatosis. Baseline clinical and histologic parameters were recorded, and compared for patients with vs. without steatosis. Multiple logistic regression analysis was performed on these baseline variables. Liver biopsies were reviewed by two pathologists, and graded for steatosis. Steatosis on biopsy was compared to steatosis on ultrasound report, and the performance characteristics of ultrasonography were calculated, using biopsy as the gold standard. **RESULTS:** Ultrasound reports were available on 164 patients. Patients with steatosis on ultrasound had a higher incidence of the following parameters compared to patients without steatosis: diabetes (12/49 [24%] vs. 7/115 [6%], $p<0.001$), fibrosis stage >2 (15/48 [31%] vs. 16/110 [15%], $p=0.02$), histologic grade >2 (19/48 [40%] vs. 17/103 [17%], $p=0.002$), and ALT (129.5 \pm 89.0 IU/L vs. 94.3 \pm 87.0 IU/L, $p=0.01$). Histologic grade was the only factor independently associated with steatosis with multivariate analysis. When compared to the histologic diagnosis of steatosis ($n=122$), ultrasonography had a substantial number of false-positive and false-negative results. In patients with a normal ultrasound, 8/82 (10%) had $>30\%$ steatosis on biopsy. Among patients with steatosis reported on ultrasound, only 12/40 (30%) had $>30\%$ steatosis on biopsy review. **CONCLUSIONS:** Steatosis on ultrasound is associated with markers of inflammation and fibrosis in HCV-infected patients, but does not consistently correlate with steatosis on biopsy outside of the context of a controlled study. Clinicians should be skeptical of the definitive diagnosis of steatosis on hepatic ultrasonography.

Additional effect of low iron diet on iron reduction therapy by phlebotomy for chronic hepatitis C. Kimura F, et al. *Hepatology*. 2005 Mar-Apr;52(6):563-6.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15816478&dopt=Abstract

BACKGROUND/AIMS: Iron-induced oxidative stress plays an important role in the pathogenesis of chronic hepatitis C. Both phlebotomy for removing body iron stores and low iron diet for minimizing portal iron supply to the liver have been shown to improve serum transaminase levels in patients with the disease. However, the cooperative effects of phlebotomy and low iron diet have not yet been elucidated in detail. **METHODOLOGY:** A pilot study was undertaken to investigate whether a low iron diet could improve the efficacy of phlebotomy in iron reduction therapy. Of 21 patients diagnosed with chronic hepatitis C, 10 patients were treated with phlebotomy alone (group A) while 11 patients were treated with a low iron plus phlebotomy (group B). Phlebotomy was repeated biweekly until serum ferritin levels reached 10 ng/mL in both A and B groups. In addition, a low iron diet (iron intake of 8 mg/day or less) was recommended for group B, followed by estimation of iron intake from daily diet records. **RESULTS:** Serum alanine aminotransferase levels were significantly improved from 106 \pm 30 to 68 \pm 22 IU/L ($p<0.005$, paired t-test) in group A and from 100 \pm 33 to 46 \pm 10 IU/L ($p<0.002$, paired t-test) in group B. The enzyme levels after treatment were significantly higher in group A ($p<0.02$, non-paired t-test), which showed a higher upward distribution of the enzyme activity. The estimated dietary iron intake in group B was reduced from 17.6 \pm 6.1 to 8.2 \pm 3.7 mg/day. **CONCLUSIONS:** These findings suggest that phlebotomy alone does not completely remove iron-induced oxidative stress and a low iron diet induces an additional effect in iron reduction therapy for chronic hepatitis C.

Treatment of chronic hepatitis C in patients unresponsive to interferon. Interest of re-treatment combining interferon induction therapy and ribavirin (a multicenter pilot study). Causse X, et al. *Gastroenterol Clin Biol*. 2005 Feb;29(2):117-21.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15795657&dopt=Abstract

AIM: About 45% of patients with chronic hepatitis C are unresponsive to the present reference treatment combining pegylated interferon plus ribavirin; before pegylated interferon was available the non-response rate was around 60%. This open multicenter pilot study, initiated before pegylated interferon became available, was designed to evaluate, in patients unresponsive to interferon monotherapy, the rate of biological and virological response and side-effects of the ribavirin-alpha 2b interferon combination. **METHODS:** The combination protocol was ribavirin (1 to 1.2 g/d) plus alpha 2b interferon at induction doses (9 MU/d the first week; 4.5 MU/d the eleven following weeks; 3 MU/2 days the 36 following weeks). **RESULTS:** Among the 27 included patients, 17 (63%) were viremia-negative (PCR) after 12 weeks of treatment, 9 (33%) were complete responders (undetectable viremia

and normal transaminases) at the end of treatment (48 weeks) and of follow-up (72 weeks). Patients with non-1, non-4 genotypes who derived full benefit from this therapeutic strategy (6/7 (86%) were complete responders: 4/5 with genotype 3 and 2/2 with genotype 5). Quality-of-life was impaired during treatment, especially during the first 12 weeks of high-dose interferon therapy. **CONCLUSION:** While waiting for new therapeutic possibilities, these good results suggest interferon induction at the beginning of treatment remains a valid option.

Prevalence and significance of neurocognitive dysfunction in hepatitis C in the absence of correlated risk factors. McAndrews MP, et al. *Hepatology*. 2005 Apr;41(4):801-8.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15793853
Neurocognitive morbidity has been reported in individuals with chronic hepatitis C virus (HCV) infection, but the magnitude of such dysfunction in the absence of disease-correlated factors known to affect the central nervous system (e.g., substance abuse, cirrhosis, depression, interferon treatment) and the impact of any such change on functioning is unclear. We investigated a cohort of individuals with HCV, all of whom were carefully screened to exclude relevant comorbidities, to elucidate virus-related changes in the brain using neuropsychological tests and magnetic resonance spectroscopy (MRS). A cohort of 37 patients with chronic HCV infection was culled from 300 consecutive patients presenting to a tertiary care liver clinic. A comparison group of healthy controls (n = 46) was also assessed. Of 10 neurocognitive measures evaluated, the HCV group showed marginally poorer learning efficiency compared with controls; only 13% of patients demonstrated a clinical level of impairment on this test (defined as 1.5 SD below the normative standard). Although patients reported greater levels of fatigue and symptoms of depression, these factors did not correlate with the degree of learning inefficiency. With respect to MRS, the HCV group demonstrated increased choline and reduced N-acetyl aspartate relative to controls in the central white matter. Indicators of liver disease severity did not correlate with either memory or MRS abnormalities. **In conclusion**, while our findings support an association between hepatitis C and indicators of central nervous system involvement in a cohort of patients carefully screened to eliminate other factors influencing neurocognitive integrity, the clinical significance of these effects is limited.

Late liver-related mortality from complications of transfusion-acquired hepatitis C. Kamitsukasa H, et al. *Hepatology*. 2005 Apr;41(4):819-25.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15793849&dopt=Abstract
Although several cohort studies have been reported in individuals with chronic hepatitis C virus (HCV) infection, little is known about liver-related mortality among the elderly. We conducted a cohort study in 302 patients with tuberculosis sequelae who had received a blood transfusion at a young age and had subsequently been treated at a chest clinic. The cohort consisted of 147 patients with antibody to HCV (anti-HCV), of whom 81% were positive for HCV RNA, and 155 without anti-HCV. The cohort was followed for a mean duration of 5.7 years. There were no differences between the two groups in the mean age of the patients at the time of transfusion (31 vs. 34 years) or at the time of entry into the study (65 vs. 66 years). The outcome of 143 patients with, and 145 without, anti-HCV could be traced; 92 (64%) and 82 (57%) had died, respectively. The main cause of death was tuberculosis sequelae in 61 (42%) and 66 (46%) patients, respectively. Eight (6%) of the 143 patients with anti-HCV died of liver disease (hepatocellular carcinoma: seven; rupture of varices: one). The average annual mortality from liver disease from study entry in the patients with anti-HCV was 9.8 per 1,000 person-years. The patients with anti-HCV had a significantly lower cause-specific survival probability for liver disease (92% vs. 100% at 10 years, P < .005). **In conclusion**, in our study, liver-related mortality appeared to be high among elderly HCV-infected individuals.

Peginterferon alfa-2b plus ribavirin treatment in children and adolescents with chronic hepatitis C. Wirth S, et al. *Hepatology*. 2005 Mar 25;41(5):1013-1018.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15793840&dopt=Abstract
Peginterferon plus ribavirin is standard therapy for adults with chronic hepatitis C. As no data are available for children, the aim of the study was to evaluate the efficacy and tolerability of peginterferon alfa-2b in combination with ribavirin in chronically infected children. Genotypes, alanine aminotransferase levels, and different routes of viral transmission were considered. In an open-labeled, uncontrolled pilot study, 62 children and adolescents (range, 2-17 years) were treated with subcutaneous peginterferon alfa-2b at a dose of 1.5 mug/kg body weight once per week plus oral ribavirin (15 mg/kg x day) for 48 weeks. Sixty-one patients completed the study. Twenty-three children discontinued therapy after 6 months according to study protocol. Sustained viral response was documented in 22 (47.8%) of 46 patients with genotype 1, in 13 (100%) of 13 with genotype 2 or 3, in 1 of 2 with genotype 4, in 19 (70.4%) of 27 children with parenteral, in 12 (48%) of 25 with vertical, and in 5 of 9 with unknown route of

infection. Overall, treatment was well tolerated. Nevertheless, some side effects were present in all treated patients. Eighty-three percent had leucopenia, but only 3 individuals required dose reduction and 10.3% developed thyroid autoantibodies and thyroid dysfunction. **In conclusion**, combination treatment of peginterferon alfa-2b with ribavirin showed encouraging results and was well tolerated in children and adolescents with chronic hepatitis C. Weekly dosing of peginterferon alfa-2b is a considerable advance for this age group. The treatment is not approved for children. Further controlled trials are needed.

Prospective evaluation of community-acquired acute-phase hepatitis C virus infection. Cox AL, et al. Clin Infect Dis. 2005 Apr 1;40(7):951-8.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15824985

BACKGROUND: More than two-thirds of hepatitis C virus (HCV) infections in Western countries are caused by injection drug use, but prospective clinical data regarding the most common mode of HCV acquisition are rare, in part because acute-phase HCV infection is usually asymptomatic. **METHODS:** To characterize acute-phase HCV infection, 179 HCV antibody-negative injection drug users were prospectively evaluated; 62 (34%) of these patients had seroconverted. Twenty of the participants who seroconverted had long-term follow-up with consistent monthly sampling before and after seroconversion, allowing detailed study. **RESULTS:** The first indication of HCV infection was the presence of HCV RNA in serum, which preceded elevation of alanine transaminase levels and total bilirubin levels to $>$ or $=2$ times baseline in 45% and 77% of patients, respectively. No subjects had jaundice. The median time from initial viremia to seroconversion was 36 days (range, 32-46 days). In one instance, viremia was detected 434 days before seroconversion. However, in no other case was HCV RNA detected >63 days before seroconversion. In subjects with viral persistence, a stable level of HCV RNA in the blood was noted in some subjects within 60 days after the initial detection of viremia, but in others, it was not apparent until >1 year later. In subjects with long-term viral clearance, HCV became persistently undetectable as early as 94 and as late as 620 days after initial viremia. **CONCLUSIONS:** These data underscore the importance of nucleic acid screening of blood donations to prevent HCV transmission and of long-term follow-up to ascertain whether there is viral persistence, at least among injection drug users.

BASIC AND APPLIED SCIENCE, PRE-CLINICAL STUDIES

Evolution of the intrahepatic T cell repertoire during chronic hepatitis C virus infection. Giuggio VM, Bonkovsky HL, Rothman AL. Viral Immunol. 2005;18(1):179-89.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15802962

Chronic hepatitis C virus (HCV) infection is characterized by extensive infiltration of inflammatory cells in the liver, where there is a compartmentalization of HCV-reactive T lymphocytes. Previous studies have demonstrated a broad intrahepatic TCR repertoire; however, there is little information regarding the stability of this intrahepatic T cell population. We studied the T cell repertoires in sequential liver biopsy samples from five individuals with chronic HCV infection using TCR spectratype analysis; four subjects had been treated with IFN-alpha during the interval between biopsies. Transcripts from most TCRBV families were detectable in the liver tissues, and 25-85% of these had skewed spectratype profiles indicative of T cell clonal expansions. Most of the intrahepatic T cell expansions were not evident in an analysis of peripheral blood T cells collected at the same time as the liver biopsy. Detailed analysis using TCRBJ-primed run-off reactions revealed that the intrahepatic TCR repertoires were not stable within an individual, although some TCR clonotypes were maintained for at least 45 months.

Transforming growth factor-beta1 gene polymorphisms are associated with progression of liver fibrosis in Caucasians with chronic hepatitis C infection. Wang H, et al. World J Gastroenterol. 2005;11(13):1929-36.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15800982

AIM: Considerable attention is focused on polymorphisms in the gene encoding transforming growth factor-beta1 (TGF-beta1), a multifunctional cytokine that is in turn a potent growth inhibitor involved in wound healing and differentiation. In humans, it promotes the pathogenesis of organ fibrosis, atherosclerosis, cancer, autoimmune and inflammatory diseases, keloid disease, and hypertrophic scarring. For this reason, much emphasis has been placed on studies elucidating the impact of TGF-beta1 and its gene variations for the susceptibility and pathogenesis of these diseases. Unfortunately, some studies have serious limitations. **METHODS:** We have recently described a high-throughput method for investigation the Arg25Pro polymorphism of human TGF-beta1 gene and showed that the frequency of the Pro25 allele is significantly associated with hepatic fibrogenesis. In this report, we describe

two novel LightCycler (LC) techniques that facilitate the examination of the two other known alterations in the coding region of TGF-beta1. We investigated whether these polymorphisms contribute to hepatitis-induced progression of fibrogenesis in Chinese and Caucasians. **RESULTS:** In the Chinese ancestry, the gene polymorphisms at codons 25 and 263 were not found and the genetic variant at codon 10 is unlikely to confer susceptibility to hepatic fibrosis. Contrarily, in Caucasians TGF-beta1 allelic variations are more frequent and the presence of prolines either in codon 25 or 10 is associated with the interindividual variability in developing more severe fibrosis during chronic hepatitis C infection. **CONCLUSION:** In summary, these results confirm the hypothesis that TGF-beta1 polymorphisms are associated with fibrosis progression in Caucasians chronically infected with hepatitis C.

Effect of hepatitis C virus core protein on cellular gene expression: specific inhibition of cyclooxygenase 2.

Jhaveri R, Kundu P, Shapiro AM, Venkatesan A, Dasgupta A. *J Infect Dis.* 2005 May 1;191(9):1498-506.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15809909

Hepatitis C virus (HCV) core protein plays a significant role in the alteration of cellular gene expression. We expressed HCV core protein using a tetracycline-inducible expression system in HeLa cell lines. Profiles of gene expression in cells expressing the HCV core protein were compared with those in control cells by use of microarray analysis. Cells expressing the HCV core protein showed 86 down-regulated and 41 up-regulated genes, compared with control cells. One gene affected was cyclooxygenase 2 (COX-2). Levels of both COX-2 RNA and the Cox-2 protein were significantly inhibited after the expression of HCV core protein in HeLa cells. Similar results were obtained in hepatoma cells and in a functional assay that measured the production of the Cox-2 protein in response to a mitogenic stimulus. The inhibition of the Cox-2 protein could serve as a means of muting the cellular inflammatory response during HCV infection. Correlation of these findings with analysis of clinical specimens from chronically infected patients should lend further significance to the down-regulation of the inflammatory response via Cox-2.

Changes of ECM and CAM gene expression profile in the cirrhotic liver after HCV infection: Analysis by cDNA expression array. Xu X, et al. *World J Gastroenterol.* 2005 Apr 14;11(14):2184-7.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15810090

AIM: We aimed to observe the expression of extracellular matrix (ECM) and cellular adhesion molecules (CAM) in cirrhotic liver tissues after hepatitis C virus (HCV) infection. **METHODS:** Twelve patients with post HCV inflammatory liver cirrhosis were selected to evaluate their liver function and other virological, pathological parameters. Then three specimens of cirrhotic patients whose health assessment results and laboratory data were similar and three normal liver specimens explanted from liver grafts prepared for liver transplantation were chosen for investigating gene expression of ECM and CAM using cDNA expression array. **RESULTS:** The cDNA array assay revealed 36.7% (36/96) of genes with changes, in which 26.3% (26/96) was up-regulated and 10.1% (10/96) was down-regulated. Integrin (ITGA), collagen (COL), ADAMTS were identified as the characteristic changes of ECM and CAM gene expression levels. ITGA were demonstrated beta1 and beta2 sub-section changed in liver cirrhosis. **CONCLUSION:** ECM and CAM play an important role in the progression of liver cirrhosis after HCV infection. The capital mechanism is related to the inflammatory cells infiltration, the activation and transformation of ECM producing cells and the imbalance between production and elimination of ECM.

Early dynamics of hepatitis C virus in the circulation of chimpanzees with experimental infection. Tanaka J, et al. *Intervirology.* 2005 Mar-Jun;48(2-3):120-3.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15812184

Two chimpanzees were inoculated with hepatitis C virus (HCV) and followed on a daily basis for 12 days. HCV RNA became detectable in their sera on day 5 by polymerase chain reaction with the detection limit of 10(2) copies/ml. Based on an exponential growth observed until 8 or 9 days after inoculation in their sera, the doubling time of HCV in the circulation was estimated at 6.3-8.6 h and log time (time required to grow 10-fold) at 31.3- 42.9 h. The exact doubling time of HCV determined in them would help plan an efficient strategy for screening out blood donors in the window period of infection between the exposure and the development of antibody to HCV in serum.

Single nucleotide polymorphisms and haplotypes in the IL10 region associated with HCV clearance. Oleksyk TK, et al. *Genes Immun.* 2005 Apr 7; [Epub ahead of print]

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15815689

Hepatitis C virus (HCV) is an infectious blood-borne pathogen that usually persists as a chronic infection. However, approximately 15% of the time, patients can clear the virus, indicating that host differences could be critical in determining the course of HCV infection. The inflammatory response is crucial to resolving or failing to resolve an acute HCV infection. Some previous reports have implicated interleukin 10 (IL10) polymorphisms with successful anti-HCV therapy and natural viral clearance. We tested 54 single nucleotide polymorphisms (SNPs) in the IL10 region (+/-300 kb and 24 within the IL10 gene itself), which contains 13 genes including the IL10 immunomodulatory paralogs IL19, IL20, and IL24, for association with HCV clearance vs persistence. SNPs from two haplotype block regions, one at IL10 and the other from IL19/IL20, were associated with HCV clearance in African Americans (91 clearance cases and 183 chronically infected matched controls; $P=0.05-0.002$) while with expectation-maximization algorithm-reconstructed haplotypes, these associations remained ($P=0.05-0.002$). However, no significant associations were detected in European Americans (108 clearance and 245 chronic). **Our results indicate** that variants of the immunomodulatory IL10 and IL19/IL20 genes may be involved in natural clearance of HCV in the African-American population.

Immunogenic Properties of a Chimeric Plant Virus Expressing a Hepatitis C Virus (HCV)-Derived Epitope: New Prospects for an HCV Vaccine. Piazzolla G, et al. *J Clin Immunol.* 2005 Mar;25(2):142-52.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15821891
A vaccine against Hepatitis C virus (HCV) is urgently needed due to the unsatisfactory clinical response to current therapies. We evaluated the immunological properties of a chimeric Cucumber mosaic virus (CMV), a plant virus engineered to express on its surface a synthetic peptide derived from many HVR1 sequences of the HCV envelope protein E2 (R9 mimotope). Evidence was obtained that the chimeric R9-CMV elicits a specific humoral response in rabbits. Furthermore, in patients with chronic HCV infection, purified preparations of R9-CMV down-modulated the lymphocyte surface density of CD3 and CD8, and induced a significant release of interferon (IFN)-gamma, interleukin (IL)-12 p70 and IL-15 by lymphomonocyte cultures. Finally, an R9 mimotope-specific CD8 T-cell response, as assessed by intracellular IFN-gamma production, was achieved in the majority of the patients studied. Our results open up new prospects for the development of effective vaccines against HCV infection. Moreover, the wide edible host range of CMV makes the production of an edible vaccine conceivable.

Hyperhomocysteinemia and the MTHFR C677T polymorphism promote steatosis and fibrosis in chronic hepatitis C patients. Adinolphi LE, et al. *Hepatology.* 2005 Apr 15;41(5):995-1003.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15834927
The factors and mechanisms implicated in the development of hepatitis C virus (HCV)-related steatosis are unknown. Hyperhomocysteinemia causes steatosis, and the methylenetetrahydrofolate reductase (MTHFR) C677T polymorphism induces hyperhomocysteinemia. We investigated the role of these factors in the development of HCV-related steatosis and in the progression of chronic hepatitis C (CHC). One hundred sixteen CHC patients were evaluated for HAI, fibrosis and steatosis grades, body mass index, HCV genotypes, HCV RNA levels, homocysteinemia, and the MTHFR C677T polymorphism. Hyperhomocysteinemia was associated with the TT genotype of MTHFR ($r = 0.367$; $P = .001$). Median values of homocysteine in the CC, CT, and TT genotypes of the MTHFR gene were 9.3, 12.2, and 18.6 $\mu\text{mol/L}$, respectively ($P = .006$). Steatosis correlated with the MTHFR polymorphism, homocysteinemia, HAI and fibrosis. Steatosis above 20% was significantly associated with fibrosis. Prevalence and high grade (>20%) of steatosis were 41% and 11% in CC, 61% and 49% in CT, and 79% and 64% in TT, respectively ($P = .01$). Relative risk of developing high levels of steatosis was 20 times higher for TT genotypes than CC genotypes. According to multivariate analysis, steatosis was independently associated with hyperhomocysteinemia (OR = 7.1), HAI (OR = 3.8), liver fibrosis (OR = 4.0), and HCV genotype 3 (OR = 4.6). On univariate analysis, fibrosis was associated with age, steatosis, MTHFR, homocysteinemia and HAI; however, on multivariate analysis, liver fibrosis was independently associated with age ($P = .03$), HAI ($P = .0001$), and steatosis ($P = .007$). **In conclusion**, a genetic background such as the MTHFR C677T polymorphism responsible for hyperhomocysteinemia plays a role in the development of higher degree of steatosis, which in turn accelerates the progression of liver fibrosis in CHC.

Upregulation of transferrin receptor 2 and ferroportin 1 mRNA in the liver of patients with chronic hepatitis C. Takeo M, et al. *J Gastroenterol Hepatol.* 2005 Apr;20(4):562-9.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15836704
Abstract Background: Iron accumulation has been reported to be associated with progression of liver injury. The mechanism of iron accumulation in the liver is not known. In the present study, hepatic messenger RNA (mRNA)

expression of transferrin receptor (TfR)1, TfR2, and ferroportin (FP)1 was measured in patients with chronic hepatitis (CH). **Methods:** Eleven patients with CH-B and 43 patients with CH-C were enrolled. All patients underwent liver biopsy. Hepatic expression of TfR1, TfR2 and FP1 mRNA was analyzed using a real-time polymerase chain reaction. Total hepatic iron score (THIS) was evaluated by Prussian blue staining. **Results:** Serum ferritin concentration is significantly higher in CH-C than in CH-B. Values of THIS of ≥ 5 were observed only in CH-C patients (44% of CH-C patients). The expression level of TfR2 mRNA was 10-26-fold higher than the TfR1 mRNA expression level. The TfR2 and FP1 mRNA expression was significantly higher in CH-C than in CH-B patients. Hepatic expression of TfR2 and FP1 mRNA was well correlated with THIS. **Conclusions:** Hepatic iron accumulation is more severe in patients with CH-C. Upregulation of hepatic iron transporters may contribute to the hepatic iron accumulation in CH-C.

Mizoribine inhibits hepatitis C virus RNA replication: Effect of combination with interferon-alpha. Naka K, et al. *Biochem Biophys Res Commun.* 2005 May 13;330(3):871-9.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15809077&dopt=Abstract
Interferon (IFN)-alpha monotherapy, as well as the more effective combination therapy of IFN-alpha and ribavirin, are currently used for patients with chronic hepatitis C caused by hepatitis C virus (HCV) infection, although the mechanisms of the antiviral effects of these reagents on HCV remain ambiguous, and side effects such as anemia due to the administration of ribavirin present a problem for patients who are advanced in years. Using a recently developed reporter assay system in which genome-length dicistronic HCV RNA encoding Renilla luciferase gene was found to replicate efficiently, we found that mizoribine, an imidazole nucleoside, inhibited HCV RNA replication. The anti-HCV activity of mizoribine (IC₅₀: approximately 100µM) was similar to that of ribavirin. Using this genome-length HCV RNA replication monitor system, we were the first to demonstrate that the combination of IFN-alpha and ribavirin exhibited more effective anti-HCV activity than the use of IFN-alpha alone. Moreover, we found that the anti-HCV activity of mizoribine in co-treatment with IFN-alpha was at least equivalent to that of ribavirin. This effect was apparent in the presence of at least 5µM mizoribine. Since mizoribine is currently used in several clinical applications and has not been associated with severe side effects, mizoribine is considered to be of potential use as a new anti-HCV reagent in combination with IFN-alpha.

Hepatitis C virus replicates in peripheral blood mononuclear cells of patients with occult hepatitis C virus infection. Castillo I, et al. *Gut.* 2005 May;54(5):682-5.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15831916&dopt=Abstract
BACKGROUND: Occult hepatitis C virus (HCV) infection is characterised by the presence of HCV-RNA in the liver in the absence of anti-HCV, and serum viral RNA. Up to 70% of these patients also have HCV-RNA in peripheral blood mononuclear cells (PBMC) but it is not known if HCV is replicating in these cells. **AIM:** We studied possible HCV replication in PBMC of 18 patients with an occult HCV infection who were selected on the basis of HCV-RNA positivity in PBMC. **METHODS:** Detection of HCV-RNA positive and negative strands in PBMC was done by strand specific reverse transcriptase-polymerase chain reaction (RT-PCR) and by in situ hybridisation. **RESULTS:** The presence of HCV-RNA positive strand in PBMC was confirmed in all patients by strand specific RT-PCR and by in situ hybridisation. Mean percentage of PBMC which had the HCV-RNA positive strand was 3.3% (95% confidence interval (CI) 2.1-4.4) The HCV-RNA negative strand was found in the PBMC of 11/18 (61%) patients by strand specific RT-PCR and confirmed by in situ hybridisation, and the percentage of PBMC harbouring the HCV-RNA negative strand was 3.1% (95% CI 0.8-5.5). There was a significant correlation ($p = 0.001$, $r = 0.84$) between the percentage of PBMC with the HCV-RNA positive strand and that of PBMC with the HCV-RNA negative strand. **CONCLUSION:** HCV replicates in the PBMC of patients with occult HCV infection and thus, although these patients do not have serum HCV-RNA, they could be potentially infectious.

Diagnosis of hepatitis C virus related acute hepatitis by serial determination of IgM anti-HCV titres. Sagnelli E, et al. *J Hepatol.* 2005 May;42(5):646-51.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15826712&dopt=Abstract
BACKGROUND/AIMS: We investigated whether serial determination of IgM to HCV core protein (HCV IgM) may be useful to identify acute hepatitis C (AHC) and to distinguish this disease from reactivation of chronic hepatitis C (r-CHC). **METHODS:** We enrolled 35 consecutive patients with AHC identified by seroconversion to anti-HCV and 31 consecutive patients who had been anti-HCV positive for at least six months at the time of reactivation. Titres of HCV IgM were calculated as Index values by a commercially available enzyme immunoassay. **RESULTS:** During the early phase of the illness we observed a wide variation in the HCV IgM

Index values in all patients in the AHC group and consistent values in all cases in the r-CHC group. The HCV viral load determined soon after the onset of symptoms was of no use in identifying AHC. Twenty-three patients in the AHC group were observed as outpatients for 6-30 months; of these, 10 became plasma HCV-RNA negative within the third month of observation, but three showed a subsequent reactivation of HCV infection. **CONCLUSIONS:** Our data indicate that the detection of high and variable titres of HCV IgM in the early phase of the illness may identify acute hepatitis C and allow early antiviral treatment.

Fibrosis correlates with a ductular reaction in hepatitis C: roles of impaired replication, progenitor cells and steatosis. Clouston AD, et al. *Hepatology*. 2005 Apr;41(4):809-18.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15793848&dopt=Abstract
The mechanisms for progressive fibrosis and exacerbation by steatosis in patients with chronic hepatitis C (HCV) are still unknown. We hypothesized that proliferative blockade in HCV-infected and steatotic hepatocytes results in the default activation of hepatic progenitor cells (HPC), capable of differentiating into both biliary and hepatocyte lineages, and that the resultant ductular reaction promotes portal fibrosis. To study this concept, 115 liver biopsy specimens from subjects with HCV were scored for steatosis, inflammation, and fibrosis. Biliary epithelium and HPC were decorated by cytokeratin 7 immunoperoxidase, and the replicative state of hepatocytes was assessed by p21 and Ki-67 immunohistochemistry. A ductular reaction at the portal interface was common. There was a highly significant correlation between the area of ductular reaction and fibrosis stage ($r = 0.453$, $P < .0001$), which remained independently associated after multivariate analysis. HPC numbers also correlated with fibrosis ($r = 0.544$, $P < .0001$) and the ductular area ($r = 0.624$, $P < .0001$). Moreover, steatosis correlated with greater HPC proliferation ($r = 0.372$, $P = .0004$) and ductular reaction ($r = 0.374$, $P < .0001$) but was not an obligate feature. Impaired hepatocyte replication by p21 expression was independently associated with HPC expansion ($P = .002$) and increased with the body mass index ($P < .001$) and lobular inflammation ($P = .005$). **In conclusion**, the strong correlation between portal fibrosis and a periportal ductular reaction with HPC expansion, the exacerbation by steatosis, and the associations with impaired hepatocyte replication suggest that an altered regeneration pathway drives the ductular reaction. We believe this triggers fibrosis at the portal tract interface. This may be a stereotyped response of importance in other chronic liver diseases.

Escape mutations alter proteasome processing of major histocompatibility complex class I-restricted epitopes in persistent hepatitis C virus infection. Kimura Y, et al. *J Virol*. 2005 Apr;79(8):4870-6.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15795272&dopt=Abstract
Mutations in hepatitis C virus (HCV) genomes facilitate escape from virus-specific CD8+ T lymphocytes in persistently infected chimpanzees. Our previous studies demonstrated that many of the amino acid substitutions in HCV epitopes prevented T-cell receptor recognition or binding to class I major histocompatibility complex molecules. Here we report that mutations within HCV epitopes also cause their destruction by changing the pattern of proteasome digestion. This mechanism of immune evasion provides further evidence of the potency of CD8+ T-cell selection pressure against HCV and should be considered when evaluating the significance of mutations in viral genomes from persistently infected chimpanzees and humans.

HIV/HCV COINFECTION

Response to interferon-based therapies in HIV-infected patients with chronic hepatitis C due to genotype 4.

Soriano V, et al. *Antivir Ther*. 2005;10(1):167-70.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15751774

BACKGROUND: The hepatitis C virus (HCV) genotype is the main predictor of response to interferon (IFN)-based therapies. HCV genotype 4 is spreading among European intravenous drug users, who are frequently coinfecting with HIV. Information about treatment response in this subset of patients is scarce and conflicting results have been reported. **METHODS:** All HIV-infected patients treated for chronic hepatitis C at our institution with a minimum follow-up of 6 months after discontinuing therapy were retrospectively analysed. They had received one of three HCV treatment modalities: IFN monotherapy, IFN plus ribavirin (RBV) or pegylated interferon (PEG-IFN) plus RBV. Treatment responses were stratified according to HCV genotype. **RESULTS:** A total of 390 patients were analysed. Sustained virological response (SVR) to HCV therapy had been reached by 90 (23.1%); 22/119 (18.5%) with IFN monotherapy; 17/106 (16%) with IFN plus RBV; and 51/165 (30.9%) with PEG-IFN plus RBV. SVR was significantly higher among those with HCV genotypes 2 or 3 (40.4%; 61/151) than in patients with either

HCV genotype 1 (11.2%; 22/197) or HCV genotype 4 (16.7%; 7/42) ($P < 0.0001$). In contrast, there were no significant differences in the response rate comparing HCV genotypes 1 and 4 ($P = 0.53$). **CONCLUSIONS:** Response to IFN-based therapies in HIV-positive patients with hepatitis C due to HCV genotype 4 is poor, similar to that obtained for HCV genotype 1 and much lower than for HCV genotypes 2 and 3. Therefore, HIV-infected patients with hepatitis C due to genotype 4 should be considered as a particular subset of difficult-to-treat patients. New treatment strategies and drugs for these patients are eagerly awaited.

Changing mortality rates and causes of death for HIV-infected individuals living in Southern Alberta, Canada from 1984 to 2003. Krentz HB, Kliwer G, Gill MJ. HIV Med. 2005 Mar;6(2):99-106.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15807715

[Canada] **OBJECTIVES:** To examine changes over a 2-year period in both the mortality rate and the causes of death in a geographically defined HIV-infected population. **METHODS:** A database search of primary care information for the dates and causes of death for all patients documented with HIV infection and living in Southern Alberta between 1984 and 2003 was undertaken. Sociodemographic and clinical characteristics were obtained. Causes of death were then individually confirmed by reviewing the patients' hospital charts, autopsy reports, or death certificates and coded using the International Classification of Diseases, 9th Revisions. AIDS deaths were reconciled with Public Health Reports. The time span was divided into pre-highly active antiretroviral therapy (HAART) (1984-1996) and current HAART (1997-2003) periods. **RESULTS:** Between 1984 and 2003, there were 560 deaths in the 1987 individuals living with HIV infection in Southern Alberta. Of these, 436 deaths (78%) occurred pre-HAART and 124 (22%) in the current HAART period. The crude mortality rate declined from 117 deaths per 1000 patient-years pre-HAART to 24 in the current HAART period. In the pre-HAART era, 90% of all deaths were AIDS related whereas only 67% were AIDS related in the current HAART era. The leading causes of AIDS deaths were AIDS multiple causes (31%), Mycobacterium avium complex (18%), Pneumocystis pneumonia (10%) and non-Hodgkin's lymphoma (7%). The proportion of non-AIDS related deaths increased from 7% pre-HAART to 32% in the current HAART era. Accidental deaths, including drug overdose (29%), suicide (7%) and violence (3%), hepatic disease (19%), non-AIDS related malignancies (19%), and cardiovascular disease (16%) accounted for the majority of non-AIDS related deaths. No deaths directly caused by drug toxicity were found. Overall, 21% of patients who died were antiretroviral (ARV)-naive. A total of 14% of patients dying from AIDS were ARV-naive in contrast to 35% dying from non-HIV related conditions. Of all those dying from AIDS, 23% died < 3 months after their initial diagnosis, reflecting late presentation. In the current HAART era, 87% of patients who died from AIDS were extensively treated, reflecting HAART treatment failures due mostly to multiclass drug resistance (42%), inexorable disease progression despite ARV (32%), lack of ability or interest to be maintained on a lifelong HAART programme (21%) and, rarely, drug intolerance (< 1%). **CONCLUSIONS:** Deaths from AIDS-related causes have decreased significantly, but deaths from non-AIDS related conditions have increased, both as an absolute number of deaths and as a proportion of all deaths in HIV-infected patients. The increasing age of the HIV population, and the increased mean CD4 count, increased proportion of intravenous drug users, increased hepatitis B virus and hepatitis C virus coinfection rate, and increased history of smoking seen in our population also influenced the mortality rate and causes of death. These factors must also be considered in projecting future trends in mortality of an HIV-infected population.

HIV coinfection shortens the survival of patients with hepatitis C virus-related decompensated cirrhosis.

Pineda JA, et al. Hepatology. 2005 Apr;41(4):779-89.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15800956

The impact of human immunodeficiency virus (HIV) coinfection on the survival of patients with hepatitis C virus (HCV)-related end-stage liver disease (ESLD) is unknown. Because HIV infection is no longer considered an absolute contraindication for liver transplantation in some countries, it has become a priority to address this topic. The objective of this study was to compare the survival of HIV-infected and HIV-uninfected patients with decompensated cirrhosis due to HCV. In a retrospective cohort study, the survival of 1,037 HCV monoinfected and 180 HCV/HIV-coinfected patients with cirrhosis after the first hepatic decompensation was analyzed. Of the group, 386 (37%) HCV-monoinfected and 100 (56%) HCV/HIV-coinfected subjects died during the follow-up. The median survival time of HIV-infected and HIV-uninfected patients was 16 and 48 months, respectively ($P < .001$). The relative risk (95% CI) of death for HIV-infected patients was 2.26 (1.51-3.38). Other independent predictors of survival were age older than 63 years (2.25 [1.53-3.31]); Child-Turcotte-Pugh class B versus class A (1.95 [1.41-2.68]) and class C versus class A (2.78 [1.66-4.70]); hepatitis D virus infection (1.56 [1.12-4.77]); model for end-stage liver disease score, (1.05 [1.01-1.11]); more than one simultaneous decompensation (1.23 [1.12-3.33]); and

the type of the first hepatic decompensation, with a poorer prognosis associated with encephalopathy compared with portal hypertensive gastrointestinal bleeding (2.03 [1.26-3.10]). **In conclusion**, HIV coinfection reduces considerably the survival of patients with HCV-related ESLD independently of other markers of poor prognosis. This fact must be taken into account to establish the adequate timing of liver transplantation in HIV-coinfected subjects.

Mitochondrial DNA depletion in liver tissue of patients infected with hepatitis C virus: contributing effect of HIV infection? Bauerle J, et al. HIV Med. 2005 Mar;6(2):135-9.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15807720

OBJECTIVES: It has been suggested that chronic hepatitis C virus (HCV) infection depletes mitochondrial DNA (mtDNA) in the liver. Because decreased mtDNA levels were also found in humans infected with HIV, we investigated whether HIV may have aggravated hepatic mtDNA depletion in individuals with HCV infection.

METHODS: In this cross-sectional study, liver biopsies were performed in a total of 40 individuals prior to any antiviral therapy. The individuals were recruited from the Hospital Clinic, Barcelona and the HIV Centre, Dusseldorf. Seventeen patients were negative for HIV and HCV and were biopsied for liver enzyme elevation of unknown cause (controls), 14 individuals had chronic HCV but no HIV infection, and nine subjects were coinfecting with both viruses. mtDNA and liver histology were centrally assessed. **RESULTS:** The groups did not differ with respect to age, gender, liver function tests and HCV viral load, where applicable. mtDNA levels were decreased by 19% in the HCV-monoinfected group (P=0.03) and by 27% in the HIV/HCV-coinfecting subjects (P=0.02) compared to controls. The mtDNA content, however, did not differ between individuals with HCV monoinfection and HCV/HIV coinfection (P=0.75). The degrees of liver fibrosis, inflammatory activity or steatosis did not correlate with mtDNA content. **CONCLUSIONS:** Liver mtDNA content is reduced in both HCV-monoinfected and HIV/HCV-coinfecting patients. Under the limitations of our study, we could demonstrate only a slight trend towards more pronounced mtDNA depletion in HIV/HCV-coinfecting subjects.

Hepatitis C augments cognitive deficits associated with HIV infection and methamphetamine. Cherner M, et al. Neurology. 2005 Apr 7; [Epub ahead of print].

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15817932

OBJECTIVE: To examine the contribution of hepatitis C virus (HCV) infection to neurocognitive dysfunction in individuals with comorbid HIV infection or methamphetamine (METH) dependence. **METHODS:** Neurocognitive functioning was examined in 430 study participants who were either normal controls or had HCV infection, HIV infection, history of METH dependence, or combinations of these factors as risks for cognitive deficits.

RESULTS: Rates of global and domain-specific neuropsychological (NP) impairment increased with the number of risk factors. HCV serostatus was a significant predictor of NP performance both globally and in the areas of learning, abstraction, and motor skills, with trends in speeded information processing and delayed recall. HCV serostatus did not predict scores in attention/working memory or verbal fluency. **CONCLUSION:** Hepatitis C virus infection contributes to the neuropsychological deficits observed among HIV-infected and stimulant-dependent populations.

HIV coinfection shortens the survival of patients with hepatitis C virus-related decompensated cirrhosis.

Pineda JA, et al. Hepatology. 2005 Apr;41(4):779-89.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15800956&dopt=Abstract

The impact of human immunodeficiency virus (HIV) coinfection on the survival of patients with hepatitis C virus (HCV)-related end-stage liver disease (ESLD) is unknown. Because HIV infection is no longer considered an absolute contraindication for liver transplantation in some countries, it has become a priority to address this topic. The objective of this study was to compare the survival of HIV-infected and HIV-uninfected patients with decompensated cirrhosis due to HCV. In a retrospective cohort study, the survival of 1,037 HCV monoinfected and 180 HCV/HIV-coinfecting patients with cirrhosis after the first hepatic decompensation was analyzed. Of the group, 386 (37%) HCV-monoinfected and 100 (56%) HCV/HIV-coinfecting subjects died during the follow-up. The median survival time of HIV-infected and HIV-uninfected patients was 16 and 48 months, respectively (P < .001). The relative risk (95% CI) of death for HIV-infected patients was 2.26 (1.51-3.38). Other independent predictors of survival were age older than 63 years (2.25 [1.53-3.31]); Child-Turcotte-Pugh class B versus class A (1.95 [1.41-2.68]) and class C versus class A (2.78 [1.66-4.70]); hepatitis D virus infection (1.56 [1.12-4.77]); model for end-stage liver disease score, (1.05 [1.01-1.11]); more than one simultaneous decompensation (1.23 [1.12-3.33]); and the type of the first hepatic decompensation, with a poorer prognosis associated with encephalopathy compared

with portal hypertensive gastrointestinal bleeding (2.03 [1.26-3.10]). **In conclusion**, HIV coinfection reduces considerably the survival of patients with HCV-related ESLD independently of other markers of poor prognosis. This fact must be taken into account to establish the adequate timing of liver transplantation in HIV-coinfected subjects.

COMPLEMENTARY AND ALTERNATIVE THERAPIES

Oral silymarin for chronic hepatitis C - a retrospective analysis comparing three dose regimens. Huber R, Futter I, Ludtke R. Eur J Med Res. 2005 Feb 28;10(2):68-70.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15817425

To investigate the effects of silymarin on aminotransferase levels in patients with chronic hepatitis C, a standardized treatment with 420mg, 840mg or 1260mg per day was performed in patients of our clinic, who were not eligible for treatment with pegylated interferon and ribavirin. Aminotransferase levels were determined before, at 3-6 week intervals during and at the end of treatment. Predefined inclusion criteria for the retrospective analysis were persistently elevated alanine aminotransferase (ALT) levels (at least 6 months prior to and at beginning of the treatment) and treatment duration of at least three weeks. Liver cirrhosis CHILD B or C, interferon therapy within the last three months before treatment with silymarin, alcohol use >30 g/d, coinfection with hepatitis B virus or other severe diseases were exclusion criteria. According to these criteria 40 patients (13 with 420mg, 20 with 840mg and 7 with 1260mg silymarin per day) were eligible for the analysis. The mean treatment period was 125 +/- 78 days. ALT, aspartate aminotransferase and gamma glutamyltransferase levels did not change significantly from baseline in any group and there were no differences between the treatment groups. Bilirubin and prothrombine time were normal in all but one patient and remained unchanged. Silymarin therapy had no side effects. Silymarin at the doses used, does not improve elevated aminotransferases in patients with chronic hepatitis C.

Oxidative stress in chronic hepatitis C: a preliminary study on the protective effects of antioxidant flavonoids. Emerit I, et al. Hepatogastroenterology. 2005 Mar-Apr;52(62):530-6.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15816472&dopt=Abstract

BACKGROUND/AIMS: Oxidative stress is involved in chronic hepatitis C, and efforts have been made to influence the disease process with antioxidants. The present study evaluates the protective effects of a phenol-rich processed grain food with superoxide-scavenging properties (trade name antioxidant biofactor AOB). **METHODOLOGY:** Thirty patients participated in this placebo-controlled double-blind pilot study. AOB was taken orally by fifteen patients for 3 mo at the recommended daily dose of 3x2 sachets, containing 3 g of powder each. Another fifteen patients received a herbal extract with practically no superoxide scavenging properties as a placebo. Oxidative stress biomarkers, aminotransferase levels and viral load were evaluated immediately before and after treatment. **RESULTS:** AOB treatment considerably improved the antioxidant defenses. Also ALT and AST decreased in 11 of the 15 patients (-11% to -65%, mean -22%, p<0.05). The effects of placebo were not significant. Viral load remained unchanged. Control biopsies were not done after the short interval of 3 mo. There were no adverse effects. After the 3-mo treatment with AOB or placebo, 16 of the 30 patients received conventional antiviral treatment (pegylated interferon alpha and ribavirin). A sustained response was observed in 5 of 9 AOB pretreated patients six mo after discontinuation of the 12-mo antiviral therapy. The 7 patients pretreated with placebo were all non-responders. **CONCLUSIONS:** These preliminary results are encouraging to conduct more extensive clinical studies combining antioxidant with antiviral treatment in hepatitis C.

MISCELLANEOUS WORKS

In situ identification of hepatitis C virus RNA in muscle. Ito H, et al. Neurology. 2005 Mar 22;64(6):1073-5.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15781833

The authors examined skeletal muscle specimens from four patients with myositis and hepatitis C virus (HCV) infection. PCR analysis identified HCV RNA in muscle homogenates from two patients. In situ hybridization signals for HCV RNA were detected within muscle fibers as well as in infiltrating lymphocytes from the same patients. The results may relate to the pathomechanism of myositis in patients with HCV infection.

Impact of hepatitis C on health related quality of life: a systematic review and quantitative assessment.

Speigel BM, et al. Hepatology. 2005 Apr;41(4):790-800.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15791608

Hepatitis C virus (HCV) diminishes health related quality of life (HRQOL), and it is now common to measure HRQOL in clinical trials. We sought to summarize the HRQOL data in HCV, and to establish the minimally clinically important difference (MCID) in HRQOL scores in HCV. We performed a systematic review to identify relevant studies, and converted HRQOL data from each study into clinically interpretable statistics. An expert panel used a modified Delphi technique to estimate the MCID in HCV. We found that patients with HCV scored lower than controls across all scales of the SF-36. Patients achieving sustained virological response (SVR) scored higher across all scales versus patients without SVR, especially in the physical health domains. HRQOL differences did not correspond with differences in liver histology or ALT levels. Based upon the published data, the expert panel concluded that the SF-36 vitality scale was most relevant in patients with HCV, and generated a mean MCID of 4.2 points on this scale. In conclusion, patients with HCV have a clinically significant decrement in HRQOL versus controls, and physical HRQOL improves in patients achieving SVR but not in those without SVR. The data further suggest that traditional outcomes fail to capture the full spectrum of illness related to chronic HCV. A difference of 4.2 points on the SF-36 vitality scale can be used as an estimate of the MCID in HCV, and this value may be used as the basis for power calculations in clinical trials evaluating HRQOL.

Estimating the impact of hepatitis C virus therapy on future liver-related morbidity, mortality and costs related to chronic hepatitis C. Buti M, et al. J Hepatol. 2005 May;42(5):639-45.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&list_uids=15826711&dopt=Abstract

BACKGROUND/AIMS: Chronic Hepatitis C virus (HCV) infection is common and often produces a progressive disease. Some studies suggest that HCV related complications will increase in the future. Our aim was to estimate the future morbidity, mortality and costs of chronic HCV infection in a cohort of patients infected by HCV and to evaluate the impact of HCV therapy. **METHODS:** A mathematical model was used to project over the next 30 years, the HCV related complications and costs in a cohort of 419,895 infected patients representing the HCV infected population in Spain. The impact of HCV therapy with peginterferon and ribavirin in this population was also projected. **RESULTS:** A gradual decline in the infected population is expected in the future, however, the proportion of patients with cirrhosis will increase by up to 14% and morbidity associated with HCV infection by up to 10% by the year 2030 with a subsequent increment in HCV related costs. However, treating from 10 to 50% of the HCV population will result in a reduction of 6 and 26% in morbidity and 4 and 20% in mortality, respectively. The cost per year of life gained ranges from 6078 for a 29-year-old patient to 8911 for a 59-year-old patient. **CONCLUSIONS:** In the future, HCV infection mortality, morbidity and associated costs will increase. Treatment of the chronic HCV infected population can eradicate the infection, increase patients' survival and reduce the need for liver transplantation, making this a cost-effective strategy.

Analytical variability of the Fibrotest proteins. Rosenthal-Allieri MA, et al. Clin Biochem. 2005 May;38(5):473-8.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15820779

OBJECTIVES: The analytical variability of the Fibrotest (FT) parameters raises the issue of the test's reliability for routine use. Whereas standardization has been proposed by the International Federation of Clinical Chemistry (IFCC) for specific proteins, few data are available concerning the actual transferability of the FT proteins, i.e. haptoglobin, apolipoprotein (A1) and alpha(2) macroglobulin. The aim of this study was to evaluate the analytical variability of the FT proteins. **DESIGN AND METHODS:** During the FIBROPACA study, we evaluated 112 sera from patients with hepatitis C infection who underwent liver biopsy. We compared measurements of haptoglobin, apolipoprotein (A1) and alpha(2) macroglobulin by the autoanalyzers Immage(R) (Beckman-Coulter) and the FT reference BNProspec(R) (Dade-Behring). **RESULTS:** Optimal concordance was found for haptoglobin (correlation: $y = 1.05x - 0.09$; correlation coefficient = 0.98). However, apolipoprotein (A1) as determined with Immage(R) was globally 12% lower than with BNProspec(R) (correlation: $y = 0.88x - 0.05$; correlation coefficient = 0.91) and alpha(2) macroglobulin values were 40% greater with Immage(R) than with BNProspec(R) (correlation: $y = 1.40x - 0.46$; correlation coefficient = 0.96). **CONCLUSIONS:** Inter-technique analytical variability of the Fibrotest parameters remains a major issue. After IFCC standardization of specific proteins, some discrepancies remain for alpha(2) macroglobulin and, to a lesser extent, for apolipoprotein (A1). National and international quality control programs would be useful to monitor analytical performance of protein assays.

Usefulness of the hepatitis C virus core antigen assay for screening of a population undergoing routine medical checkup. Gaudy C. et al. J Clin Microbiol. 2005 Apr;43(4):1722-6.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15814991

We studied the usefulness of the recently designed Trak-C assay for the detection and quantification of the hepatitis C virus (HCV) core antigen (Ag) for the screening of HCV infection in 4,201 subjects selected from 74,150 consecutive volunteers undergoing routine medical checkups. Subjects were selected for screening because they had risk factors (group II, n = 321) and/or elevated alanine transaminase activity (group I, n = 3847). Initially, the anti-HCV antibody assay and the Trak-C assay were performed on each patient. Subsequently, the Trak-C assay was performed only when the anti-HCV enzyme immune assay (EIA) was positive. Positive samples were further evaluated for anti-HCV antibodies by a third-generation strip immunoblot assay and for HCV RNA. Four samples (1.2%) from group II and 113 (2.9%) from group I were anti-HCV EIA positive. We also tested 33 subjects who previously tested positive for anti-HCV in our medical center. Among the 150 anti-HCV EIA-positive samples, the HCV core Ag result was in accord with the HCV RNA result in 146 cases (97.3%). When the EIA result was positive, the HCV core Ag concentration and the HCV RNA load were correlated ($r(2) = 0.78$; $P < 0.001$). Four samples with low viral loads were Trak-C negative but HCV RNA positive. Among the 2,395 anti-HCV EIA-negative serum samples collected during the first part of the study, 17 (0.7%) were found to contain very low levels of HCV core Ag (< 8.5 pg/ml, the cutoff value being 1.5 pg/ml). All these samples were HCV RNA negative and considered to be false positives. This was confirmed by HCV core Ag neutralization analysis. The HCV core Ag assay is a useful method in the screening strategy of HCV infection and provides a reliable means of distinguishing between current and cleared HCV infections that is well correlated with HCV RNA testing.

Biopsychosocial predictors of fatigue in chronic hepatitis C. Hilsabeck RC, Hassanein TI, Perry W. J Psychosom Res. 2005 Feb;58(2):173-178.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15820845

OBJECTIVE: The objective of this study was to investigate the relationship of biopsychosocial variables to fatigue in patients with chronic hepatitis C (CHC) with the goal of identifying areas of possible intervention.

METHOD: Ninety-four patients with CHC not on antiviral therapy completed a demographic questionnaire and measures of fatigue, health-related quality of life, and depression. Fibrosis stage and relevant medical information were obtained from medical records. **RESULTS:** A stepwise multiple regression analysis revealed that the most significant predictor of fatigue was poor social functioning, followed by poor physical functioning, greater depression, and female gender. Together, these variables accounted for 68% of the total variance in fatigue.

DISCUSSION: Study findings suggest that treatment strategies for fatigue in patients with CHC should focus on increasing social functioning in addition to improving physical functioning and decreasing depressive symptoms. Research investigating the effectiveness of interventions targeting social functioning, such as support groups and supportive counseling, is recommended.

Sexual transmission of HCV between spouses. Tahan V, et al. Am J Gastroenterol. 2005 Apr;100(4):821-4.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15784025

[Turkey] **BACKGROUND AND AIMS:** The sexual transmission of hepatitis C virus (HCV) is debated. By excluding other risk factors, the role of sexual intercourse in the transmission could be detected more accurately. We screened HCV prevalence and risk factors in the spouses of chronic hepatitis C (CHC) patients and followed the seroconversion rate of anti-HCV negative spouses. **PATIENTS AND METHODS:** Six hundred spouses of CHC patients were recruited. The spouses' HCV risk factors were questioned and the spouses were tested for anti-HCV. The 216 spouses who were anti-HCV negative were checked annually for anti-HCV. **RESULTS:** Anti-HCV was positive in 12 of 600 (2%) of the spouses. Of the 12 anti-HCV positive spouses, 11 were HCV-RNA positive. Of anti-HCV positive and negative spouse groups, mean age was 52.3 +/- 9.8 and 49.8 +/- 12.4 yr; duration of marriage was 1521 +/- 506.7 and 1532.4 +/- 670.2 wk ($p > 0.05$); and the number of total sexual intercourse was 434 +/- 295 and 307 +/- 333 ($p = 0.055$), respectively. In our prospective study, none of the spouses developed anti-HCV seroconversion in mean 35.7 +/- 6.3 months and 257.9 +/- 72.2 sexual intercourse. **CONCLUSIONS:** Anti-HCV was found positive in 2% of the spouses. None of the seronegative spouses developed seroconversion in the 3-yr follow-up period. This is the first study that stresses the importance of the total number of sexual intercourse in sexual transmission ($p = 0.055$). Our results of special monogamous group with very limited risk factors support the role of number of total sexual intercourse in HCV transmission. However, the seroprevalence rate of the spouses was still within the upper limit of our country population.

The hepatitis C crisis. St John TM, Sandt L. Ethn Dis. 2005 Spring;15(2 Suppl 2):S52-7.

http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=pubmed&dopt=Abstract&list_uids=15822840

An estimated 170 million persons (3% of the world's population) are infected with the hepatitis C virus (HCV), and 3 to 4 million persons are newly infected each year. Of those infected, 70%-85% develop chronic viremia with the potential for devastating long-term sequelae, including chronic liver disease, cirrhosis, hepatic failure, and hepatocellular carcinoma. The passivity in the public health sector and in the medical community at large with respect to hepatitis C portends a myriad of societal, fiscal, and personal costs for the United States within the next two decades unless immediate actions are taken to intervene in the natural history of this emerging public health crisis.

ABSTRACTS OF INTEREST: 40TH ANNUAL MEETING EASL, APR. 13-17, 2005. PARIS, FR.

NOTE: Abstracts of the meeting can be searched for and viewed at <http://www.kenes.com/easl2005sci/program/search.asp>

Modeling the natural course of chronic hepatitis C: validation and clinical implications. Veldt BJ, et al. Abstract 615.

Background/Aims: The outcomes of cohort studies describing the natural course of chronic hepatitis C are variable. Models estimating the risk of clinical events may be unreliable if validation with real patient data is lacking. Furthermore, realistic estimates of survival should also take non-liver-related mortality into account.

Methods: We developed a Markov model using rates of fibrosis progression derived from a large histological study in patients with a known duration of infection, and rates of progression to clinical events, derived from a clinical study in cirrhotics. Outcomes were adjusted for non liver-related mortality and tested against published cohort studies identified by systematic review of the literature. **Results:** The absolute risk of liver-related mortality within 30 years after infection was 8.5% (95% CI 5.3-11.8) for males and 5.5% (95% CI 4.7-6.5) for females infected at 30 years of age. The standard mortality ratio (SMR) over time increased only up to 2. If cirrhosis was present, the SMR increased more markedly: for cirrhotic patients aged thirty, the SMRs at 10-year follow-up were 24.6 for males and 36.8 for females. Although disease progression was faster in older patients, the SMR remained below 2 due to a simultaneous increase in non-liver-related mortality. The absolute risk of liver related mortality within 30 years after infection was 28.3% (95% CI 21.9-30.8) for males and 20.6% (95% CI 15.1-26.8) for females infected at 50 years. The outcomes of the model matched the results of published sizeable cohort studies closely.

Conclusions: By integrating rates of fibrosis progression and longitudinal data in cirrhosis, the present model provides realistic estimates for the long-term clinical outcome of chronic hepatitis C. This model may help identifying those patients most at risk of excess mortality from hepatitis C infection.

Long-term progression of fibrosis in patients with chronic hepatitis C (CHC) non-responders to interferon therapy (IFN). A longitudinal study of repeat liver biopsies. Cirera I, et al. Abstract 555.

Aims: To assess the rate of fibrosis progression in CHC patient's non-responders to IFN and to identify associated variables. **Patients and methods:** 84 patients (50.5±1.3 years) with CHC non-responders to IFN, and without cirrhosis in their initial biopsy, followed for 8.1±0.2 years. In 67 of them a second liver biopsy was performed after 8.6±1.7 years. In the remaining 17 cases the second biopsy was not performed because the diagnosis of cirrhosis was established after hepatic decompensation (10) or hepatocellular carcinoma (7). Liver biopsy samples were re-examined blindly by a pathologist who was unaware of the clinical and biochemical data of the patients or the order of the biopsies. Studies on the liver biopsies included assessment of fibrosis (METAVIR), and of steatosis. **Results:** In the basal biopsy, fibrosis was F0 in 19 patients, F1 in 19, F2 in 10 and F3 in 36. Steatosis was absent in 40 cases, but was grade I in 36 and grade II in 8 cases. The overall rate of fibrosis progression was 0.104 units per year. Fibrosis progression was found in 52 patients (62%) (13/19 patients with F0 in the basal biopsy, 11/19 F1, 6/10 F2, and 22/36 F3). Variables associated with fibrosis progression were age, HCV-RNA, GGTP, AP, AST/ALT, platelets, cholesterol and steatosis in the initial biopsy. The independent predictors for fibrosis progression were platelet count (OR 0.09; 95%IC: 0.01-0.67) and steatosis (OR 4.96; 5%IC: 1.11-25.5). 27 patients (32%) developed cirrhosis (1 with F0 in the basal biopsy, 3 F1, 1 F2, and 22 F3), and 11 patients (13%) developed hepatocellular carcinoma. **Conclusions:** 1) The results of this longitudinal study have shown progression of liver fibrosis, cirrhosis and hepatocellular carcinoma development in approximately one half, one third and one tenth, respectively, of CHC patients non-responders to IFN in a median follow-up of 8 years. 2) The variables independently associated with fibrosis progression were thrombocytopenia and steatosis in the initial biopsy. 3) Re-treatment is highly indicated in this subgroup of patients with high risk of fibrosis progression.

Virologic response and safety outcomes in therapy-naïve patients treated for chronic hepatitis C with viramidine in combination with pegylated interferon alfa-2a. Gish RG, et al. Abstract 91.

Introduction: Dose-limiting anemia can be a prominent adverse event of therapy with pegylated interferon and ribavirin. This dose-ranging study examined whether viramidine, a liver-targeting prodrug of ribavirin, may be a safer alternative when used in combination with pegylated interferon alfa-2a (PEG-IFN). **Methods:** Of 180 HCV therapy-naïve patients enrolled in the study, 171 patients received full-dose viramidine (400 mg: n = 47; 600 mg: n = 43; 800 mg: n = 44) versus ribavirin 1000-1200 mg/d (n = 37) in combination with PEG-IFN 180 µg/wk SC. Patients were predominantly male (64%), Caucasian (76%), and genotype 1 (72%), with a median HCV RNA of 6.5 log₁₀ copies/mL. Analyses assessed the incidence of anemia (hemoglobin <10 g/dL) and HCV RNA levels (Bayer TMA assay; sensitivity to 5 IU/mL) among patients without dose reduction due to anemia to evaluate the intrinsic activity of viramidine versus ribavirin without the confounder of dose modification. **Results:** Among patients with no dose modification due to anemia at end of treatment (EOT), no significant differences were noted between viramidine (400, 600, and 800 mg BID) versus ribavirin in the proportion of patients with undetectable HCV RNA levels (55%, 63%, 55%, and 62%, respectively). Rates of anemia at EOT for the viramidine 400, 600, and 800 mg groups were 0%, 2%, and 11%, respectively, versus 27% for the ribavirin arm. Based on evaluable patients at EOT experiencing a decline in hemoglobin of at least 25%, the rate in the ribavirin group (48%) was higher versus the rate in the viramidine groups (400 mg: 14%; 600 mg: 18%; 800 mg: 15%). Other types of adverse events were similar between treatment arms. **Conclusions:** At EOT in this Phase 2 study, viramidine demonstrated antiviral activity comparable to that of ribavirin when used in combination with pegylated interferon alfa-2a among patients with no dose modification due to anemia. Patients in the viramidine arms also showed a significantly lower incidence of anemia.

Tolerability and immunogenicity of HCV E1 therapeutic vaccination in patients with chronic hepatitis C on interferon plus ribavirin. Horsmans Y, et al. Abstract 568.

Background: In patients with chronic hepatitis C, open-label E1 therapeutic vaccination monotherapy has been shown to be well tolerated, immunogenic, and associated with improvement in liver histology. **Aim:** This prospective open-label study was conducted to explore the immunogenicity and tolerability of E1 therapeutic vaccination in patients who had already started standard interferon/ribavirin therapy for at least 10 weeks. **Methods:** Seventeen patients were enrolled; one patient was ineligible. Sixteen patients with chronic hepatitis C (10 male, 6 female; mean age 46.4 years; genotype 1 or 3; median duration of IFN-based therapy 15 weeks (range 10-37 weeks); 11 previously treatment-naïve) received 6 intramuscular injections of 20 µg HCV E1 at three-week intervals with a 4-week follow-up. T-cell responses were measured by proliferation assays, cytokines and anti-E1 antibodies by ELISA. Quality of life (QoL) was assessed with the RAND 36 questionnaire before and after E1 vaccination. **Results:** HCV E1 induced both E1-specific humoral and cellular immune responses. The median values of anti-E1 antibody levels increased from 87 mU/ml at baseline to 185 at study end. Of the 10 patients tested, nine developed a positive T cell response (SI ≥ 3) with a strong increase in IFN-γ and IL-5 secretion. With the exception of local side effects at the site of vaccine injection, no unusual side effects were observed. Interestingly, a trend towards improvement was observed for 4 out of 8 QoL scales and for Health Transition. Of the nine patients with an elevated ALT value at the start of vaccination, three normalized serum ALT levels. Of the 10 patients who still had detectable viremia at the start of vaccination, 3 became negative during combined treatment. **Conclusion:** A course of 6 HCV E1 injections was well tolerated and was associated with an E1-specific immune response as well as a trend towards improvement in QoL in patients with chronic hepatitis C who already received at least 10 weeks of standard therapy. Some “late” biochemical and viral responses were observed. Whether such responses can be induced by the addition of HCV E1 therapeutic vaccination to standard therapy remains to be investigated in larger controlled studies.

Association between liver histology, viral kinetics and outcome of therapy with peginterferon alfa-2a combination therapy with ribavirin in interferon naïve patients with chronic hepatitis C. Lagging M, et al. Abstract 574.

Background and aims: Previous studies have shown that the presence of liver cirrhosis is associated with lower sustained viral response rates after Peginterferon-ribavirin therapy. The aim of this study was to evaluate the association between liver histology, viral kinetics, and treatment outcome after Peginterferon-ribavirin combination therapy even for milder degrees of liver fibrosis. **Methods:** 272 interferon naïve patients with chronic hepatitis C genotype 1-5 viral infection underwent therapy with Peginterferon alfa-2a 40KD in combination with ribavirin in

accordance with the DITTO-HCV multi-center trial. 231 of these patients had a liver biopsy that could be evaluated centrally in a blinded fashion by two independent observers using the Ishak protocol. Equivocal issues were debated after the independent scores were noted, and a consensus score was obtained. HCV-RNA levels were determined centrally using the Roche AMPLICOR HCV Test. **Results:** Liver fibrosis was associated with viral kinetics during the first 4 weeks of therapy with null responders having statistically higher fibrosis stages (median stage 3 vs. stage 2 for responders; $p=0.0083$, Mann-Whitney U-test). Likewise having the presence bridging fibrosis (Ishak stage 3) and not just cirrhosis was associated with a lower SVR rate in genotype 1 (38% vs. 61%; $p=0.027$, Mann-Whitney U-test). A similar trend for lower SVR when bridging fibrosis was present was also noted for genotype 2 (83% vs. 100%), genotype 3 (86% vs. 92%), and genotype 4 (0% vs. 50%) though these differences were not statistically significant. No association was seen between fibrosis stage and baseline viral load. However, patients having an inflammation sum score (i.e. Histological Activity Index) had statistically higher viral loads at baseline (mean 6.47 vs. 6.09 log IU/mL; $p=0.045$, Mann-Whitney U-test). **Conclusion:** Liver histology is associated with baseline viral load prior to, viral kinetics during, and sustained virological response after therapy with Peginterferon and ribavirin. Thus assessment of liver histology remains pivotal in decision-making regarding therapeutic intervention especially in the presence of HCV genotype 1 infection.

Are current fibrosis serum markers really an alternative to liver biopsy in chronic hepatitis C? Leroy V, et al. Abstract 577.

We recently described a score of fibrosis combining PIIINP and MMP-1, which are involved in fibrogenesis and fibrolysis respectively. The aim of this study was to evaluate its diagnostic accuracy compared to that of a panel of other markers including prothrombin time (PT), hyaluronate (HA), Fibrotest (FT), APRI and Forns' score in chronic hepatitis C (CHC) patients **Methods:** One hundred and eighty CHC patients seen in our centre for a pre-therapeutic liver biopsy were prospectively included. Liver fibrosis was assessed using the METAVIR index. Sinusoidal fibrosis, steatosis and iron load were quantified. Biochemical measurements were performed on serums collected the day of the biopsy. **Results:** Median length of liver biopsies was 22 mm. Patients were classified as F0:n=15, F1:n=74, F2:n=40, F3:n=26, F4:n=24. Overall diagnostic accuracy of the 4 combined scores was better than that observed with PT and HA alone. AUROC ranged from 0.79 to 0.84 for discriminating F0/F1 vs F2/F3/F4 and from 0.81 to 0.88 for discriminating F0/F1/F2 vs F3/F4 (NS). Accuracy was not altered by the length of biopsies or by the presence of sinusoidal fibrosis, steatosis or iron in liver. Applying the lower cut-off described for each score, METAVIR fibrosis greater than F1 could be ruled out in about one third of patients with NPV ranging from 82 to 84%. The higher cut-off selected about 20% of patients whom extensive fibrosis (F3F4) was confirmed with a PPV ranging from 89 to 93%. Simultaneous use of at least 2 scores improved NPV up to 100% for the diagnosis of F0F1, but decreased the number of selected patients. Commercial application of FT proposes an estimated fibrosis METAVIR. Discordances of at least 1 point were observed in 52% and of 2 points in 22% of patients. The only parameter associated with discordance was FT between 0.20 and 0.80. **Conclusion:** Our results confirm that PIIINP/MMP-1 provides relevant information on liver fibrosis with a good accuracy, comparable to that observed with FT, APRI and Forns' score. However none of these scores can reliably give and estimated METAVIR index in each patient. Combination of several scores could improve their accuracy.

Peginterferon alfa-2a versus peginterferon alfa 2b in the treatment of chronic hepatitis C. Mauss S, et al. Abstract 585.

Background: Recent studies in HCV-monoinfected patients may suggest differences in the efficacy or tolerance of the two currently approved pegylated interferons. Prospective, controlled studies are ongoing, but data will not be available before 2006. **Methods:** To gain controlled, comparative data we conducted a matched pair analysis for patients treated with peginterferon alfa-2a or peginterferon alfa-2b from our data base ($n=427$). Eligible were all patients with first interferon-based therapy and follow-up since initiation of therapy for at least 18 months for hcv-genotype 1 ($n=90$) or 4 ($n=4$) and 12 months for genotype 2 ($n=18$) or 3 ($n=54$). Patients were matched within each center for hcv-genotype and hcv-rna ($< vs. >800.000$ IU/mL). Therapy consisted of peginterferon alfa-2a (180 μ g qw) or peginterferon alfa-2b (1.5 μ g/kg qw) plus ribavirin 800-1200 mg bid based on weight. Primary endpoint is hcv-rna <100 IU/ml 24 weeks post treatment (SVR). For statistical analysis Mann Whitney and Fisher test was used. At baseline patients treated with peginterferon alfa-2a had a median age of 39 years, and median hcv-rna of 453000 IU/mL, not statistically different from peginterferon alfa-2b 40 years and hcv-rna 489000 IU/mL. Body mass index tended to be lower in the peginterferon alfa-2a group, 23.5 kg/m² vs. 24.9 kg/m² ($p=0.06$). **Results:** Sustained virologic response (SVR) and relapse after end of treatment response (EOTR) are shown in the table. Premature discontinuation due to adverse events or patient request was identical with 27/83 patients (32%) each.

Conclusion: In this matched pair analysis no statistical significant differences in efficacy and tolerance were observed between peginterferon alfa-2a and peginterferon alfa-2b.

Differential TH1/TH2 cytokine profile of chronic hepatitis C patients with sustained virological response compared to non-responders and the effect of IFN plus ribavirin treatment. Par1 G, et al. Abstract 451.

Background: T-helper 1 (Th1) cytokines are required for host antiviral cell-mediated immunity, Th2 cytokines are responsible for humoral immune response and may associate with persistence of virus. The aim of the study was to compare Th1 and Th2 type cytokine production of stimulated T cells and macrophages in patients with chronic hepatitis C having different responses to antiviral therapy. **Methods:** 28 patients with chronic hepatitis C undergoing IFN+ribavirin therapy and 20 healthy controls have been studied. Th2 (IL-4, IL-6, IL-10) and Th1 (IFN γ , TNF α , IL-2) cytokine levels were determined by FACS microbead assay from the supernatant of LPS stimulated macrophages and PMA+ionomycin stimulated T cells before the onset of antiviral treatment and at 1, 3, 6 month thereafter. **Results:** Pretreatment IFN γ production of the T cells was significantly decreased in both non-responders (NR) (151 \pm 18 pg/l) and sustained responders (SR) (94 \pm 27 pg/l) compared to healthy controls (206 \pm 22pg/l). Pretreatment capacity of the macrophages to produce Th1 type cytokines and IL-6 was significantly higher in patients who became later sustained responders (IFN γ : 493 \pm 25 pg/l, TNF: 4,23 \pm 1,1, IL-6: 87 \pm 26) compared to the non-responders (IFN γ : 9 \pm 0,7pg/l, TNF: 1,02 \pm 0,2pg/l, IL-6: 26 \pm 5pg/l) and healthy controls (IFN γ : 11 \pm 0,1pg/l, TNF: 0,52 \pm 0,1pg/l, IL-6: 21 \pm 2pg/l). Although antiviral treatment downregulated the production of IL-6, IFN γ and TNF α by peripheral macrophages, it increased IL-2 and IFN γ production by T cells in the responders. IL-10 production was significantly higher in non-responders (1220 \pm 24 pg/l) compared to sustained responders (560 \pm 14 pg/l) and healthy controls (710 \pm 11 pg/l). **Conclusion:** In chronic hepatitis C patients with sustained virological response the increased production of Th1 type cytokines by the macrophages may counteract impaired Th1 cytokine production of T cells and may play a crucial role in sustained virological response. Non-responders showed a Th2 type cytokine profile. Antiviral treatment increased Th1 type cytokine levels in sustained responders that may be important in inducing cellular immune response against HCV infection.

Clustering of poor prognostic factors in patients with chronic hepatitis C. Swain M, et al. Abstract 607.

Body weight is a modifiable factor that influences the outcome of interferon-based therapy for chronic hepatitis C. Sustained virological response (SVR) rates tend to decrease as body weight increases. The mechanism whereby body weight affects the response to treatment is not completely understood, but is likely to be multifactorial. We assessed the complex relationship between body weight and a range of baseline host- and hepatitis C virus (HCV)-related factors that influence the outcome of interferon-based therapy. **Methods:** Baseline data from two multinational, randomized, controlled phase III studies assessing the efficacy and safety of peg-IFN α -2a (40KD)/ribavirin for the treatment of hepatitis C were combined (NEJM 2002;347:975; Ann Intern Med 2004;140:346). Patients were classified into 3 groups: <65 kg, 65–<85 kg, and \geq 85 kg. Baseline patient- and HCV-related characteristics were analyzed according to body weight. **Results(table):** Data from a total of 2404 patients were included in the analysis. Body weight was positively correlated with body mass index (BMI), the proportion of male patients, the proportion of black patients, those reported to have acquired HCV through intravenous drug use (IVDU), and with a histological diagnosis of cirrhosis. A greater proportion of patients infected with HCV genotype 1 were in the highest weight group (\geq 85 kg) than in the other lower weight groups, although this was not statistically significant. There was no apparent correlation between body weight and ALT levels. **Conclusions:** Body weight has a complex relationship with a range of patient- and disease-related characteristics. In general, patients in the highest weight group were much more likely to be male, black, to have cirrhosis and to have been infected through IVDU. This clustering of poor prognostic characteristics may explain the lower SVR rates observed in heavier patients.

Induction of the TH1/TC1 type immunity in chronic hepatitis C non-responder patients with the therapeutic peptide vaccine IC41. Wedemeyer H, et al. Abstract 19.

IC41 is a vaccine consisting of five synthetic peptides harboring relevant CD4+ and CD8+ T-cell epitopes of Hepatitis C Virus (HCV) and the synthetic Th1/Tc1 adjuvant poly-L-Arginine. IC41 has been investigated in a number of clinical trials including up to date about 240 subjects confirming both its excellent safety profile and its immunogenicity. Recently, results of a double blind, controlled, randomized, multicenter phase-2 study in chronic hepatitis C patients who did not respond to standard interferon-based therapies were presented (Manns et al., AASLD 2004). IC41 treatment involved 6 s.c. vaccinations in 4-weekly intervals. To investigate the mode-of-action of IC41 in chronically infected patients more specifically, we now performed a detailed analysis of

frequencies, kinetics and phenotypes of T cell responses induced by IC41. Peripheral blood mononuclear cells (PBMC) were isolated and cryo-preserved at 8 time points before, during and after IC41-vaccination. T-cell responses were assessed with a combination of standardized assays in one central laboratory: interferon-gamma ELISpot, lympho-proliferation and HLA-tetramer analysis. 21 out of 36 patients (58%) treated in the verum groups showed a T-cell response to IC41-vaccination. Both, interferon-gamma secreting CD4+ helper and CD8+ cytotoxic T-cells (CTL) were induced by IC41. Importantly, Poly-L-Arginine was required to induce this type of response. Overall, the strength of IC-41-peptide-specific CD4+ and CD8+ T cell responses increased during treatment, peaked 4 weeks after the last vaccination and declined thereafter. A significant decline of HCV RNA levels during vaccination was observed in 6 of 21 patients with a T-cell response. 3 of those achieved a greater than 1-log decrease which was however transient. Interestingly, these patients showed a strong and sustained reactivity to one particular CTL-epitope. CD8+ T-cells recognizing this epitope as well as non-HCV control peptides were characterized in detail involving staining for surface markers (CD8, CD45RA, CCR7, CD27, CD28, CD38, CD69 and HLA-DR) and intracellular perforin revealing significant differences between HCV and CMV- or EBV-specific CTLs. **In summary**, cellular immunity against HCV can be enhanced by therapeutic vaccination with IC41. These data could be the basis for novel treatment strategies in chronic HCV infection, either as monotherapy or in combination antiviral therapies.

Evidence that serum ribavirin steady-state concentrations do not correlate with early virological response during peginterferon alfa-2a + ribavirin combination therapy for chronic hepatitis C. Souvignet C, et al. Abstract 606.

Background/aims: Ribavirin in combination with pegylated interferon is the standard treatment for chronic hepatitis C. Despite recommendations for ribavirin dosage based on body weight of the patient, serum ribavirin concentrations display high inter-individual variability that could explain severe side effects or lack of efficacy in some patients. The present study aimed to define the relation between serum ribavirin concentration and viral load drop on combination treatment. **Methods:** Thirty-six patients chronically infected with the hepatitis C virus (genotypes 1 or 4) were treated with standard treatment (180 µg Peginterferon alfa-2a once weekly and 1000-1200 mg ribavirin twice daily). During the first three months of treatment, the serum ribavirin concentration was sequentially monitored using a HPLC specific assay, in parallel with measures of viral load (Roche Amplicor™ HCV test, v2.0) and haemoglobin level at the same time points (Days 0, 7 and 14, and Weeks 4 and 12). **Results:** The steady-state concentration of ribavirin was reached at Week 4: 1.99 ± 0.73 mg/ml (Variation coefficient = 41 %). Mean drop of viral load was -2.26 ± 0.97 log UI/ml at Week 4 and -2.93 ± 0.77 log UI/ml at Week 12. Undetectable HCV-RNA was noted in 36 percent of the patients at Week 4 and 83 percent at Week 12. No significant correlation was found between serum ribavirin concentration (Week 4) and viral load drop (Week 12): $r = 0.071$ ($p = 0.679$). The ribavirin concentration that gave an early virological response in 50 percent of the patients (ED50) was 2.17 mg/ml. The ribavirin concentration that gave 2 g/dL drop of haemoglobin in 50 percent of the patients (TD50) was 2.44 mg/ml at the same timepoint (Week 12). **Conclusions:** During the first three months of combination treatment for chronic hepatitis C (Peginterferon alfa-2a + ribavirine), the drop of viral load does not correlate with serum ribavirin concentration. These data suggest that interindividual variations of serum ribavirin levels do not significantly impact early virological response rate. Actually, the interest of therapeutic drug monitoring of ribavirin will depend on further analysis of sustained virologic response data after long-term follow-up of this patient cohort.

Peginterferon alfa 2-b therapy in acute hepatitis C: impact of onset and duration of therapy on sustained virologic response. Kamal S, et al. Abstract 15.

BACKGROUND: The role, onset and duration of peginterferon treatment have not been evaluated in acute hepatitis C. **OBJECTIVE:** To assess the efficacy, safety, onset and duration of peginterferon alfa (PEG IFN) in acute hepatitis C. **METHODS:** In this intent to treat study patients with acute HCV genotypes 1 and 4 (n=98) were enrolled and prospectively followed. Patients were screened for 8 weeks after seroconversion or first positive PCR. Fifteen subjects refused treatment but were followed through the study. Patients without spontaneous recovery were randomized to begin PEG IFN-alpha monotherapy at wks 8, 12, 20 respectively for either 12 or 24 weeks. A subset of subjects who failed to achieve a virologic response after 12 wks of treatment continued therapy for additional 12 wks. **RESULTS:** Five untreated subjects had spontaneous recovery and another 4 subjects scheduled to start treatment at weeks 12 or 20 resolved spontaneously before therapy. 79 subjects with persistent viremia were randomized to 3 groups (Table). The end of treatment response was 94% and the overall SVR was 82%. The SVR was better for genotype 4 compared to genotype 1. Earlier treatment (week 8 or 12) was associated with higher

SVR particularly in genotype 1. Twelve week therapy was sufficient for genotype 4 while higher SVR rates in genotype 1 patients were achieved with 24 wks treatment (86%). Peginterferon alfa-2b monotherapy was well tolerated and associated with significant improvement in the quality of life. **Conclusion:** Peginterferon alfa-2b monotherapy improves sustained virologic response for acute hepatitis C virus with genotype 1 and 4 infection. Earlier treatment leads to increased virologic response. HCV genotype 1 may require longer treatment duration.

Fibrosis assessment in hepatitis C by computer-performed optical analysis of CT of the liver: a novel non-invasive and useful method. Romero-Gomez M, et al. Abstract 599.

Background: Computer-performed optical analysis of images from conventional CT without contrast media is a novel, non-invasive, reproducible method to assess the distribution and staging of fibrosis in the whole liver of patients with chronic hepatitis C (CHC). **Purpose:** To validate this tool in a cohort of 90 patients suffering from biopsy-proven CHC. **Patients and Methods:** Ninety patients (60/30=men/women, age=42+10 years) were enrolled. Histological fibrosis was staged following Scheuer F-score: F0-F1=45(50%), F2=19(21.1%) and F3-F4=26(28.8%). AST/platelets ratio (APRI) and Forns Index were calculated. Conventional helical CT axial slices without contrast media (120 kV, 190-205 mAs, 5 mm/7 mm=thickness/intersection gap) were obtained and digitized (0.20 mm/pixel, 8 bpp). Window limits (Level=Width=71) were selected by an expert radiologist and kept constant for all images. Digital analysis comprised calculation of physical parameters describing histogram features (=optical spectra, OS) on a sampling grid overlaid to CT scans. OS-Weighted-Mean-of-Fibrosis (OS-WMF) was calculated as a non-linear weighted combination of samples with each calculated-F score. **Results:** OS-WMF correlates with fibrosis ($r=0.67$; $p<0.001$), increasing with F-stage: F0=0.77±1.08; F1=1.19±1.14; F2=1.64±1.99; F3=2.88±0.61; F4=3.34±0.36 (ANOVA, $p<0.0001$). Mean fibrosis of the cohort was 1.7±1.3 by histopathological analyses and 1.8±1.3 by OS-WMF, $p=ns$. Since OS-WMF was 1.27±1.13 in F0-F2 and 3.09±0.55 in F3-F4, $p<0.0001$, it discriminates patients with F3-F4: if OS-WMF<2, advanced fibrosis can be excluded (NPV=97.9%) and if OS-WMF>3 the possibility of showing F3-F4 is high (PPV=90%). However, one third of patients with fibrosis F0-F1 showed OS-WMF>2. **Conclusions:** Computer-performed optical analysis of images from conventional CT of the liver is a reliable method, able to measure hepatic fibrosis in patients with hepatitis C. Weighted mean value and distribution of fibrosis in the liver can be quantified. Major discrepancies with histopathological results are found in patients with higher fibrosis stage. Whether this is a bias of proposed method or an error due to sampling limitations of liver biopsy requires further analysis.

Does the clinical outcome of hepatitis C infection vary with the infecting hepatitis C virus type? Harris HE, et al. Abstract 567.

It is not known whether differences in the natural history of hepatitis C virus (HCV) can be explained by differences in the infecting genotype. The aim of this study was to describe the prevalent genotypes in a national cohort of patients who acquired HCV infection at a known date and to investigate whether there was any evidence to suggest that the natural history of their infections might vary with the infecting type. Data on clinical outcomes were extracted from the HCV National Register and mortality data were taken from death certification. Sera were available for 749 cases who had been enrolled in the UK HCV National Register. HCV RNA-positive specimens were genotyped and HCV RNA-negative specimens were serotyped. Logistic regression analysis was used to investigate the effect of HCV type on viral clearance and histological stage of liver disease. 444 of the sera were found to carry HCV RNA. The most prevalent HCV types were 1 (52%), 3 (29%) and 2 (16%). Of the 305 specimens found to be HCV RNA-negative, it was possible to serotype 160 of them; serotypes 1 (58%), 3 (24%) and 2 (13%) were the most common. There was no evidence of any association between viral type and: mortality, signs and symptoms of liver disease or histological grade of liver disease. A significant association was observed between viral type and response to treatment, with type 1 infections being associated with poor response to treatment when compared to types 2 or 3 ($P=0.003$). Viral type was independently associated with spontaneous viral clearance, with type 1 being more likely to clear spontaneously than non-1 types (OR 0.50, 95% CI 0.30–0.84, $P=0.009$). Viral type was also independently associated with histological stage of liver disease, with type 1 being associated with stage scores above the median when compared to non-1 types (OR 2.03; 95% CI 1.07–3.83; $P=0.03$). **The results of this study suggest** that HCV type 1 infection is more likely to be associated with spontaneous clearance but is clinically more aggressive than type non-1 infection when it persists.

Characteristics associated with progression of fibrosis in women with hepatitis C: evidence for the role of estrogen. Codes L, et al. Abstract 556.

Background and aims : The rates of fibrosis progression in chronic hepatitis C are significantly different between males and females. An antifibrogenic effect of estrogen is proposed, probably via the inhibition of stellate cells. The aim of this study was to evaluate the evolution of hepatitis C in women, according to use of oral contraceptives, number of pregnancies, menopause and substitutive hormonal therapy. **Methods :** From November 2001 to October 2002, women with chronic hepatitis C were enrolled prospectively. Patients without liver histology or with other cause of liver dysfunction were excluded. A questionnaire was completed prospectively and a blood sample was obtained on the day of liver biopsy. Liver biopsies were evaluated according to the METAVIR. We identified predictive factors of liver fibrosis by using univariate and multivariate (logistic regression) analysis. **Results :** Our study included 251 women. Average age was 46.7 +/- 10.8 years-old, duration of the infection was 24.2 +/- 6.7 years. Pregnancy was distributed as follows : 24.8% no pregnancies, 56.4% with 1 or 2 pregnancies and 18.8% with 3 or more pregnancies, 50.8% had antecedents of use of contraceptives. A total of 122 patients (52.1%) were menopausal and 65 received substitutive hormonal therapy. We didn't identify relationship between number of pregnancies, use of contraceptives and fibrosis. Patients with a high stage of fibrosis (F2-F4) were older, had a longer duration of infection, a higher BMI. They presented liver steatosis more frequently and received hormonal therapy less frequently than patients with low stage fibrosis (F0-F1). Among 65 menopausal women treated with substitutive hormonal therapy, 53 presented fibrosis F0-F1. When we analyzed all of patients that received hormones (contraceptive pills or replacement therapy), we found a predominance of treated women (86.3%) in the group F0-F1 ($p < 0.0001$). A high stage of fibrosis was associated with higher AST levels [OR 3.5 (95% CI 1.5-8.2), $p=0.0038$] and previous anti-viral therapy [OR 2.8 (1.2-6.6), $p=0.0171$]. The probability of high stage fibrosis was lower in the group of patients who had received hormonal therapy [OR 0.26 (95% CI 0.11-0.62), $p=0.002$]. **Conclusions :** These results reinforce the hypothesis of a probable antifibrogenic role of estrogens.

Enhanced antiviral efficacy for valopicitabine (NM283) plus peginterferon in hepatitis C patients with HCV genotype-1 infection: results of a phase 2a multicenter trial. Afdhal N, et al. Abstract 93.

Background: Only 40-50% of HCV-1 infected hepatitis C patients respond to pegylated interferon (peg-IFN α) plus ribavirin therapy. NM283 is a novel nucleoside analog that as monotherapy reduced serum HCV RNA by a mean 1.2 log₁₀ (94%) in a recent 15-day trial in HCV-1 patients, most of whom had previously failed antiviral therapy for HCV infection. NM107, the parent compound of NM283, and interferon alfa exhibit synergistic antiviral effects against BVDV models in vitro, prompting the current investigation of NM283 combined with peg-IFN α in treatment naïve patients with chronic hepatitis C. **Methods:** A multicenter, open-label phase IIa trial is evaluating whether NM283 plus peg-IFN α -2b has enhanced antiviral activity compared to NM283 alone in HCV-1 infected patients. Key entry criteria are: HCV RNA >5 log₁₀ IU/mL, ALT <5 xULN, compensated liver disease, treatment naïve. Eligible patients are randomized 2:3 to NM283 or peg-IFN α + NM283 (combinationRx). NM283 is dosed orally QD for 24 weeks in both groups, escalating to 800 mg/day over the first week and then continuing at that dose. The group randomized to combinationRx receives peg-IFN α -2b (1.0 μ g/kg weekly) starting on Day 8. **Results:** Presently 19 of 30 planned patients (age 22-65, 68% males) are enrolled and have received 10 or 12 weeks of treatment. Tolerance of both treatments has been satisfactory. Mean HCV RNA reductions (log₁₀ IU/mL) from baseline to the last patient visit are 1.0 for the NM283 monotherapy group and 3.2 for the combinationRx group. Eleven of twelve combinationRx patients treated for at least 10 weeks have had substantial HCV RNA reductions (range 1.2-6.2 log₁₀), and 8 of 12 patients have achieved >2 log₁₀ decrease in HCV RNA, which has been associated with sustained response to current standard Rx. Presently 4 patients are PCR-negative (<10 IU/mL). 12-week early virological response data will be presented at the meeting. **Conclusions:** NM283 combined with peg-IFN α shows consistent, rapid and marked anti-HCV activity in patients with HCV-1 infection, a historically difficult-to-treat group. Initial EVR rates with combinationRx are encouraging and support continued investigation of NM283 and NM283 + peg-IFN α , which may offer improved efficacy and tolerability for patients with HCV-1 infection.

Accurate models predicting response to pegylated interferon with ribavirin in patients with chronic hepatitis C as early as week 4. Martinot-Peignoux M, et al. Abstract 584.

Current models used to predict response to peginterferon plus ribavirin treatment, based on viral decline during the first 12 weeks of therapy, have focused on creating an early stopping rule to avoid unnecessary prolongation of therapy. **Methods.** We developed a multivariate model that predicted sustained virological response (SVR) and non-response (NR) at baseline, and during the first 12 weeks of therapy using collected data from 186 unselected patients with chronic hepatitis C treated with peginterferon plus ribavirin. These models employed ordinal regression with similarity least squares technology to assign the probability of a given outcome. Model variables

included sex, age, prior treatment status, genotype, baseline serum alanine aminotransferase levels, histologic necroinflammation and fibrosis scores and serum hepatitis C virus RNA concentration at baseline, weeks, 4, 8, and 12. **Results.** Multivariate models demonstrated high performance values at all time points. The multivariate baseline model demonstrated negative predictive value (NPV) and positive predictive value (PPV) of 91% and 95%, respectively and for the week 4 model these values had improved to 97% and 100%, respectively with 95% sensitivity, 89% specificity and 93% accuracy. The week 4 model was equally efficient for naive or previously treated patients. Its internal validation demonstrated 90% PPV, 94% NPV, 95% sensitivity, 88% specificity and 92% accuracy. **Conclusion.** A week 4 stopping rule for patients with chronic hepatitis C treated with Pegylated Interferon with Ribavirin might be proposed by using the model developed in our study.

Prediction of sustained virological response at week 4 of peginterferon alfa-2a plus ribavirin treatment in chronic hepatitis C patients with persistently normal ALT levels. Tran A, et al. Abstract 612.

In general, the absence of an early virological response (EVR) at week 12 of combination therapy with peginterferon alfa-2a (40KD) plus ribavirin (RBV) has high negative predictive value (NPV) for the probability of achieving a sustained virological response (SVR). We aimed to determine whether this criterion could be applied at week 4 of treatment. **Methods:** 422 patients with persistently normal ALT levels (≤ 30 IU/L, on ≥ 3 occasions over 18 months) were randomized to 24 or 48 weeks of treatment with peginterferon alfa-2a (40KD) 180 μ g/week + RBV 800 mg/day. The study was conceived before pivotal studies revealed that RBV 1000/1200 mg/day is optimal for genotype 1. SVR was defined as undetectable HCV RNA (< 50 IU/mL) after 24 weeks of follow-up. Rapid virological response (RVR) was defined as undetectable HCV RNA or a ≥ 2 -log decline in HCV RNA at week 4. **Results** (TABLE): The NPV of an RVR in genotype 1 patients treated for 48 weeks (79%) is lower than that of an EVR at week 12 in the same group (96%, J Hepatol 2004;40(Suppl):48). However, when only patients with genotype 1 and a high viral load (HVL) are considered, the NPV is 100% at week 4. The NPV of an RVR was 50% in the overall genotype 2/3 patient population. **Conclusions:** The results show that general treatment decisions should not be based on an RVR at week 4 in patients treated with peginterferon alfa-2a (40KD) plus RBV. The high NPV of an RVR at week 4 in genotype 1, HVL patients is intriguing. Extending treatment duration to 72 weeks in patients without an RVR (predominantly genotype 1, $\approx 40\%$ HVL) significantly improved SVR rates in a recent trial (TeraViC-4, J Hepatol 2004;40(suppl 1):150). Thus, further study is required to determine the optimal duration of treatment for genotype 1 patients without an RVR at week 4.

| Patients (N) | With a rapid virological response at wk 4 (%): | With an SVR at wk 72 (%): | PPV (%) | NPV (%) |
|---|--|---------------------------|---------|---------|
| All genotype 1 pts treated x 48 wk (141) | 61/141 (43) | 57/141 (40) | 66 | 79 |
| Genotype 1 HVL treated x 48 wk (51) | 25/51 (49) | 14/51 (27) | 56 | 100 |
| All genotype 2,3 pts treated x 24 or 48 wks (117) | 109/117 (93) | 88/117 (75) | 77 | 50 |
| Genotype 2,3 HVL treated x 24 or 48 wks (56) | 51/56 (91) | 39/56 (70) | 71 | 40 |

HVL = high viral load = HCV RNA $> 800,000$ IU/mL ; PPV = positive predictive value (probability of an SVR after an RVR); NPV = negative predictive value (probability of no SVR without an RVR)

Is delivery mode related to perinatal transmission of hepatitis C virus (HCV). Mariné- Barjoan E, et al. Abstract 582.

Objective: to analyse factors related to perinatal HCV transmission. **Methods :** Multi-centre prospective study conducted in HCV-positive mothers in Southern France (Alpes-Maritimes, Haute-Garonne, Hérault) between October 1998 and September 2002. Mothers' and children's clinical, HCV/HIV virological characteristics and mode of delivery were recorded. All babies were controlled at M3 by PCR-RNA. They were considered infected if the HCV-RNA was positive with the same genotype as the mother's. **Results :** The analysis was conducted in 240 children. HCV-RNA was positive in 136 mothers and 26% were HIV-positive. Eighty children were delivered by caesarian section, 80% before onset of labour. In 63% of cases membranes were intact. Fifty-six percent of these were elective caesarian sections. Twelve children were HCV-infected (5.6%) at 12 months' follow-up, among whom 3 HCV-RNA became negative at 18 months. Their mothers all had positive HCV-RNA at the time of delivery. Rate of transmission was significantly higher among HIV/HCV co-infected mothers compared with HCV-infected mothers (11.0% versus 3.8 %; $p=0.05$). None of the children were HIV-infected. There was no statistically significant association between HCV transmission and mode of delivery (5 contaminated children among 45 deliveries by elective caesarian (11%) versus 7 contaminated children born by vaginal delivery (4%), timing of caesarian section and condition of the membranes. Among HCV-RNA positive mothers, the risk of HCV transmission was significantly higher for women with a high viral load (median 5.99 log copies/ml versus 5.68 log copies/ml, $p < 0,008$). After adjusting for study centre, the risk was higher among HIV-positive women (OR aj: 4.6

95%CI 1.006-21.734). **Conclusions:** A high HCV viral load at the time of delivery and HIV co-infection are the main risk factors for perinatal transmission of HCV. Caesarian section, whether elective or not, is not associated with a lower risk of HCV transmission. Spontaneous resolution of viral RNA is possible in children. .

Development and validation of a simple model to predict response to peginterferon plus ribavirin combination therapy in genotype 1 chronic hepatitis C. Martínez-Bauer EM, et al. Abstract 583.

The aim of this study was to develop simple scoring indexes to predict virological response prior to or early during pegylated interferon plus ribavirin combination therapy in naïve, HCV genotype 1 infected patients with chronic hepatitis C. **Methods:** Two indexes, a baseline predictive index (BPI) and a week 4 of therapy predictive index (W4PI) were constructed using data from a cohort of 104 patients consecutively treated with pegylated interferon alfa-2b and ribavirin at a single hospital. The validity of BPI and W4PI was assessed in an external cohort of 141 comparable patients recruited and identically treated at four independent hospitals. Virological response was defined by negative HCV-RNA at the end of therapy and 24 weeks after treatment withdrawal. No response was defined by persistence of HCV-RNA after 24 weeks of therapy or by relapse. **Results:** Serum HCV-RNA concentration, AST:ALT ratio, serum cholesterol, and a calculated non-invasive score aimed to estimate liver fibrosis were independently associated to response by uni and multivariate analysis of baseline data in the estimation cohort. At week four of therapy, BPI and clearance of HCV-RNA were independently associated to response. BPI and W4PI were formulated using these variables and the power of these indexes to predict response was explored by receiving operator (ROC) curves. The area under the ROC curve in the estimation group and in the validation group were, respectively, 0.856 and 0.847 for BPI, and 0.908 and 0.907 for W4PI. In the estimation cohort, a BPI score lower than 6.60 had 97% sensitivity and 92% negative predictive value and a score higher than 9.70 had 96% specificity and 91% positive predictive value. A W4PI score lower than 1.35 had 97% sensitivity and 93% negative predictive value and a score higher than 6.02 had 98% specificity and 96% positive predictive value. The predictive values of BPI and W4PI in the validation group were closely similar. **In conclusion,** BPI and W4PI may be valuable for therapeutic decision and to design studies for evaluation of alternative therapeutic strategies in genotype 1-infected patients with chronic hepatitis.

Effect of maintenance peg-intron therapy on portal hypertension and its complications: results from the COPILOT study. Curry M, et al. Abstract 95.

The COPILOT clinical trial is evaluating maintenance therapy with PEG-Intron 0.5mcg/kg weekly versus colchicine (COLC) 0.6mg twice daily in 600 patients with HCV and advanced fibrosis who have failed prior interferon treatment. Clinical endpoints include death, liver failure, transplantation, variceal bleeding and liver cancer. The 2 year evaluation suggested an improved event free survival in patients on PEG compared to colchicine (Hepatology 2004;40;239A). We evaluated the role of portal hypertension (PHTN) on the clinical outcomes of maintenance therapy and the role of maintenance PEG-Intron on portal pressure. **PATIENTS / METHODS:** 537 patients are enrolled in the trial to date; 267 COLC and 270 PEG. 83% of patients in each group have cirrhosis and 40% have PHTN defined by varices or gastropathy. Endoscopy is performed every 2 years for development of new varices. A substudy evaluated portal pressure using HVPG measurements prior to PEG and after 24 weeks of PEG in 5 patients with varices who were not on B blockers. Clinical endpoints were evaluated in patients with and without PHTN. **RESULTS:** 132 patients have had repeat endoscopy at 2 years and new varices were seen in 11 of 66 patients on COLC versus 5 of 66 patients on PEG (p =ns). All 5 patients had baseline elevated HVPG (Mean 15mmHg) and all had a reduction in HVPG after 24 weeks on PEG (Mean 6 mmHg; overall HVPG reduction 41%,mean reduction in HVPG 7mmHg). Primary event rate for patients with PHTN was 13.5% per year on COLC and 5.5% per year on PEG (p < 0.004). Primary event rate without PHTN was 3% in each treatment group. Variceal bleeding over 2 years occurred in 11 patients on COLC (9%) and 1 patient on PEG (1%). Ascites and liver failure was also more common in COLC (n = 20) compared to the group on PEG (n=13). **CONCLUSION:** Maintenance PEG therapy may retard varices development, reduces portal pressure and prevents variceal bleeding and complications of PHTN compared to COLC. Clinical endpoints are more common in patients with PHTN. PEG should be considered in patients with cirrhosis who fail interferon and ribavirin therapy.

Modulation of interferon specific gene expression by albuferon™ in subjects with chronic hepatitis C and correlation with antiviral response. Moore P, et al. Abstract 447.

Background: Albuferon™ is a novel recombinant protein derived from IFNalpha genetically fused to human serum albumin. The resulting single polypeptide combines in one molecule the antiviral properties of IFNalpha with the long serum half-life of albumin. As interferon mediated biological responses stem from the engagement of IFN

with its target receptor and subsequent modulation of Interferon specific gene (ISG) expression, gene expression response were evaluated and compared with anti-viral response in subjects with chronic hepatitis C treated with Albuferon. **Methods:** Whole blood was obtained at multiple time points during the Phase 1/2 study conducted in subjects with HCV who had previously failed IFNalpha containing regimens. Subjects were enrolled sequentially to receive 1 or 2 subcutaneous injections of Albuferon up to 900 mcg. Gene expression was determined by a Taqman real-time PCR assay and DNA array analysis. **Results:** cDNA array analysis identified several ISG's in subjects treated with Albuferon or pegylated IFN. Real-time PCR analysis of candidate ISG expression in 21 subjects enrolled in higher dose Albuferon (400-900 mcg) cohorts demonstrated modulation of interferon specific gene responses. There was sustained induction of the ISG's- OAS1, IRF-7, IFI44, and IFI27, up to 1000 fold over a 28-day period. Temporal regulation of Granulysin (serine protease) expression and down-regulation of DcR2 (regulator of apoptosis) expression was also observed. While all subjects showed a molecular response to Albuferon, individual variability in pre-treatment gene expression levels and fold of modulation during treatment was observed. The response to Albuferon assessed either by real-time PCR or DNA array analysis was similar to that observed previously with both pegylated IFN's. Regression analysis showed a positive correlation ($p < 0.05$) between induction of IFI44 and OAS1 gene expression and antiviral response at Day 28. Furthermore, there was a dose dependent increase in the magnitude and durability of OAS1 induction at Day 28 over baseline following a single injection when the lower dose cohorts (7-80 mcg) and higher dose cohorts (400-900 mcg) were compared. **Conclusions:** Albuferon demonstrated robust induction of ISG that was consistent with the response associated with an IFNalpha.