

Spontaneous mutation of K-ras in adult human stem cells would be the equivalent developmental path in sporadic pancreatic cancer. Depending on when this occurs, a small or large island of clonal K-ras mutant cells would develop other mutations and epigenetic events within this population could give PanIN lesions and eventually invasive carcinoma. A carcinoma originating in a small field of mutant cells will be likely to have developed earlier in the progression of the K-ras clones than a carcinoma in a large field; the consequence of this for the patient are unclear but it is at least possible that a longer period of preneoplastic development would give a greater chance of selecting mutations that in the context of a cancer cell will give greater invasiveness.

If the paper by Kim and colleagues¹ does indicate that patients with fields of cancerisation will have a better prognosis, then two types of patient will be defined: those with tumours more amenable to treatment and those in whom tumours will develop in a large potentially identifiable region of molecular changes. In the latter case, early identification of risk on the basis of molecular tests may allow early

detection, and nothing in Kim's paper contradicts the notion that early detection will also equate to good prognosis.

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REFERENCES

- 1 Kim J, Reber HA, Dry SM, *et al.* Unfavourable prognosis associated with K-ras gene mutation in pancreatic cancer surgical margins. *Gut* 2006;**55**:1598–1605.
- 2 Neoptolemos JP, Stocken DD, Dunn JA, *et al.* The influence of resection margins on survival for patients with pancreatic cancer treated by adjuvant chemoradiation and/or chemotherapy within the ESPAC-1 randomized controlled trial. *Ann Surg* 2001;**234**:758–68.
- 3 Karpoff HM, Klimstra DS, Brennan MF, *et al.* Results of total pancreatectomy for adenocarcinoma of the pancreas. *Arch Surg* 2001;**136**:44–7.
- 4 Bogoevski D, Yekebas EF, Schurr P, *et al.* Mode of spread in the early phase of lymphatic metastasis in pancreatic ductal adenocarcinoma: prognostic

significance of nodal microinvolvement. *Ann Surg* 2004;**240**:993–1000.

- 5 Brennan JA, Mao L, Hruban RH, *et al.* Molecular assessment of histopathological staging in squamous-cell carcinoma of the head and neck. *N Engl J Med* 1995;**332**:429–35.
- 6 Braakhuis BJ, Tabor MP, Kummer JA, *et al.* A genetic explanation of Slaughter's concept of field cancerization: evidence and clinical implications. *Cancer Res* 2003;**63**:1727–30.
- 7 Seymour AB, Hruban RH, Redston M, *et al.* Allelotype of pancreatic adenocarcinoma. *Cancer Res* 1994;**54**:2761–4.
- 8 Paulino AC. Resected pancreatic cancer treated with adjuvant radiotherapy with or without 5-fluorouracil: treatment results and patterns of failure. *Am J Clin Oncol* 1999;**22**:489–94.
- 9 Neoptolemos JP, Stocken DD, Friess H, *et al.* A randomized trial of chemoradiotherapy and chemotherapy after resection of pancreatic cancer. *N Engl J Med* 2004;**350**:1200–10.
- 10 Shimizu Y, Yasui K, Matsueda K, *et al.* Small carcinoma of the pancreas is curable: New computed tomography finding, pathological study and postoperative results from a single institute. *J Gastroenterol Hepatol* 2005;**20**:1591–4.
- 11 Yan L, McFaul C, Howes N, *et al.* Molecular analysis to detect pancreatic ductal adenocarcinoma in high-risk groups. *Gastroenterology* 2005;**128**:2124–30.
- 12 Tuveson DA, Shaw AT, Willis NA, *et al.* Endogenous oncogenic K-ras (G12D) stimulates proliferation and widespread neoplastic and developmental defects. *Cancer Cell* 2004;**5**:375–87.
- 13 Tuveson DA, Zhu L, Gopinathan A, *et al.* Mist1-KrasG12D knock-in mice develop mixed differentiation metastatic exocrine pancreatic carcinoma and hepatocellular carcinoma. *Cancer Res* 2006;**66**:242–7.

Hepatitis C

Altered monoaminergic transporter binding in hepatitis C related cerebral dysfunction: a neuroimmunological condition?

D M Forton

There may be a role for disturbed monoaminergic neurotransmission in the pathophysiology of hepatitis C virus associated cerebral dysfunction

Fatigue, depression, and complaints of mild cognitive impairment, such as poor concentration and forgetfulness, are the commonest symptoms reported by patients with chronic hepatitis C virus (HCV) infection.¹ Yet there remains considerable debate as to whether these symptoms are caused by the virus itself. Fatigue is a multidimensional symptom with multiple, sometimes coexisting, determinants which may be biological, psychological, or sociological. It is an

important cause of impaired health related quality of life (HRQL) in HCV infection.² Numerous surveys have documented high prevalences of fatigue but consistently show no relationship with the degree of liver fibrosis, markers of inflammation, or viral load.³ This has led to the conclusion that there is no causal relationship between HCV and neuropsychological symptoms.⁴ Rather, psychological processes associated with diagnostic labelling, social functioning, anxiety about treatment, substance

abuse, and depression have been invoked to account for impairments in HRQL.^{5,6} In contrast, a number of neuroimaging studies, including a single photon emission computerised tomography (SPECT) study published in this issue by Weissenborn and colleagues,⁷ have suggested that measurable abnormalities exist within the central nervous system (CNS) in a proportion of HCV infected individuals (*see page 1624*).^{8–11}

The issue has tended to become polarised between functional and biological arguments, and the likely interaction between physical and psychological factors has been relatively ignored. Attempts have been made to control for relevant confounding factors to determine whether CNS dysfunction relates directly to HCV infection. In a carefully executed study where 300 HCV infected patients were screened for potential risk factors for cognitive impairment such as cirrhosis, psychiatric comorbidity, or previous substance abuse, a highly selected cohort of only 37 patients was identified to have no likely cause for cerebral dysfunction, other than HCV infection itself.¹¹ This small group underwent cognitive testing and patients were found to have significant impairments in learning efficiency, which did not relate to fatigue and depression, which were also

reported. These findings followed on from previous studies which had demonstrated deficits in attention, learning ability, and memory in HCV infected individuals without cirrhosis.^{9–10, 12}

Cerebral magnetic resonance spectroscopy gives information on cerebral metabolism and has been used to test the hypothesis that a biological mechanism underlies the neuropsychological dysfunction in HCV infection. Four published studies have showed significant alterations in cerebral choline (Cho) and N-acetylaspartate (NAA) in HCV infected patients without cirrhosis.^{8–11} The findings of elevated Cho and reduced NAA mirror those reported in human immunodeficiency virus (HIV) infection,¹³ a virus which is tropic to the CNS. Detection of replicative intermediates of HCV (negative strand RNA) within the CNS¹⁴ and different viral variants in the CNS, liver, and serum¹⁵ support the concept of low level HCV replication within the brain. Although the mild neurocognitive impairments seen in HCV infection are not progressive as in AIDS dementia, it has been suggested that they may result from cerebral immune activation, possibly as a result of CNS infection by HCV.⁹

There is some clinical evidence that ondansetron, a serotonin type 3 receptor antagonist, may ameliorate HCV associated fatigue.¹⁶ In view of this and the evidence of cognitive and cerebral metabolic abnormalities in HCV infection, Weissenborn and colleagues sought to determine whether altered monoaminergic neurotransmission is associated with cognitive dysfunction in selected patients.⁷ They studied 20 patients with exposure to HCV, 16 of whom were still viraemic and four who had no detectable virus in serum, as determined by polymerase chain reaction (PCR). Patients had been referred to a tertiary hospital neurology clinic for assessment of fatigue and cognitive decline. In agreement with previous studies, these patients displayed varying degrees of neurocognitive impairment, predominantly in the domain of attention. They also recorded high rates of depression, anxiety, and fatigue. The four PCR negative patients appeared to be equally impaired on all scales. Patients were studied with SPECT to measure serotonin and dopamine transporter binding capacity (SERT and DAT, respectively). Statistically significant reductions in hypothalamus/midbrain SERT and striatal DAT binding were found compared with healthy controls. Pathological SERT and DAT binding were evident in 50–60% of HCV exposed cases, including three of the four PCR negative patients. There were no correlations between the

SPECT data and fatigue, mood, or HRQL. However, patients with impaired DAT or DAT and SERT binding did worse as a group on the cognitive tests compared with both healthy controls and HCV infected patients with normal SPECT measurements. These novel findings are interpreted as implicating a role for disturbed monoaminergic neurotransmission in the pathophysiology of HCV-associated cerebral dysfunction.

A number of lines of less direct evidence support this conclusion. The therapeutic use of cytokines such as interleukin 2 (IL-2) and interferon α (IFN- α) is associated with the induction of depressive symptoms in patients with cancer or viral hepatitis.¹⁷ These symptoms respond to treatment with the selective serotonin reuptake inhibitors, which are active at the presynaptic serotonin transporter.¹⁸ This has led to research into the immune basis of depression by investigators within the psychoneuroimmunological community.¹⁹

Interactions between the immune system and serotonergic neurotransmission have been demonstrated at a number of levels, both peripherally and within the CNS. Cytokine receptors are expressed on glia and neurones within the brain. Peripherally derived cytokines may signal to the CNS through a number of pathways,²⁰ including induction of proinflammatory cytokines by perivascular macrophage-like cells, saturable transport across the blood brain barrier at high concentrations, and an action on afferent nerves to the CNS such as the vagus nerve.²¹ This mechanism may be particularly relevant in HCV infection where the cytokine milieu in the liver, innervated by the vagus nerve, is deranged. Although the basal level of cytokine production within the brain is likely to be low, a network exists whereby cells, particularly microglia, may produce cytokines in response to peripheral signals. For example, peripheral administration of lipopolysaccharide to rats results in intracerebral IL-1 β production.²² Cerebral immune activation may alter the metabolism of key monoamines (for example, IL-1 β increases expression of the serotonin transporter gene *in vitro*).²³ There is also evidence that IFN- α increases serotonin uptake *in vitro* through increased expression of the serotonin transporter,²⁴ and that intracerebroventricular injections of IFN- α in rats reduce frontal cortex and midbrain serotonin concentrations in a dose dependent manner.²⁵

Studies of IFN- α administration in humans have generated data on serotonin metabolism. IFN- α increases serum kynurenine (KYN) concentrations and

reduces serum serotonin and tryptophan (TRP) concentrations and these changes have been shown to correlate with depression ratings.^{17–26} The mechanism whereby this occurs is thought to be related to induction by IFN- α of the enzyme indoleamine 2,3-dioxygenase (IDO), expressed on immune cells, including microglia.²⁷ IDO catalyses the conversion of TRP to KYN, reducing the availability of TRP for serotonin synthesis. There is also evidence that endogenous cytokine production in states of chronic immune activation, such as HIV infection or rheumatoid arthritis, may result in TRP depletion and high KYN levels, expressed as an increased KYN/TRP ratio.²⁸ For example, in HIV infection, elevated KYN/TRP correlates with levels of IFN- γ and neopterin,²⁹ suggesting that in states of chronic Th-1 type immune activation, IDO is induced. Furthermore, an association between TRP depletion and cognitive impairment has been reported in HIV infection.³⁰ To date, there are no data in chronic HCV infection but a similar interaction seems possible.

There are therefore a number of possible mechanisms through which peripheral and central immune activation could result in alterations in monoaminergic neurotransmission in HCV infection. These mechanisms remain theoretical and untested in HCV infection. Given the complexity of these systems and the possibility of changes in regulation of monoaminergic transporters and receptors over time in chronic disease, the functional significance of the findings of reduced mid-brain SERT and striatal DAT binding in this study remains unclear. Although the role of these brain regions in cognitive processing is not resolved, the findings in this study do implicate a role, or at least an association, between disturbed monoamine function and cognitive function in HCV infection. Indeed, the concept of cerebral immune activation as the basis for these changes may allow a model that incorporates both the biological and psychological theories to date. Animal data suggest that psychogenic stressors and proinflammatory cytokines may result in similar outcomes, in terms of neurotransmitter activity.²⁰ This may go some way to explaining why reduced SERT and DAT binding were observed in three of the four patients who had cleared HCV from serum. As Weissenborn and colleagues⁷ have postulated, there may be a sustained CNS effect even after the virus has been eradicated from serum, which may result in some form of sensitisation, conferring increased vulnerability to psychogenic stressors.

CNS symptoms are only present in a proportion of individual with HCV infection. In others it is a truly asymptomatic condition. It is likely that these symptoms result as a consequence of a complex interplay between viral and host genetic factors and external stressor events. Further investigation of the CNS effects of HCV infection and chronic immune activation may enable, in time, the development of strategies to treat the neuropsychological symptoms in those who do not respond to or tolerate antiviral therapy.

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REFERENCES

- 1 Poynard T, Cacoub P, Ratziu V, et al. Fatigue in patients with chronic hepatitis C. *J Viral Hepat* 2002;**9**:295–303.
- 2 Kramer L, Hofer H, Bauer E, et al. Relative impact of fatigue and subclinical cognitive brain dysfunction on health-related quality of life in chronic hepatitis C infection. *AIDS* 2005;**19**(suppl 3):S85–92.
- 3 Goh J, Coughlan B, Quinn J, et al. Fatigue does not correlate with the degree of hepatitis or the presence of autoimmune disorders in chronic hepatitis C infection. *Eur J Gastroenterol Hepatol* 1999;**11**:833–8.
- 4 Wessely S, Pariente C. Fatigue, depression and chronic hepatitis C infection. *Psychol Med* 2002;**32**:1–10.
- 5 Barrett S, Goh J, Coughlan B, et al. The natural course of hepatitis C virus infection after 22 years in a unique homogenous cohort: spontaneous viral clearance and chronic HCV infection. *Gut* 2001;**49**:423–30.
- 6 Rodger AJ, Jolley D, Thompson SC, et al. The impact of diagnosis of hepatitis C virus on quality of life. *Hepatology* 1999;**30**:1299–301.
- 7 Weissenborn K, Ennen JC, Bokemeyer M, et al. Monoaminergic neurotransmission is altered in hepatitis C virus infected patients with chronic fatigue and cognitive impairment. *Gut* 2006;**55**:1624–30.
- 8 Forton DM, Allsop JM, Main J, et al. Evidence for a cerebral effect of the hepatitis C virus. *Lancet* 2001;**358**:38–9.
- 9 Forton DM, Thomas HC, Murphy CA, et al. Hepatitis C and cognitive impairment in a cohort of patients with mild liver disease. *Hepatology* 2002;**35**:433–9.
- 10 Weissenborn K, Krause J, Bokemeyer M, et al. Hepatitis C virus infection affects the brain—evidence from psychometric studies and magnetic resonance spectroscopy. *J Hepatol* 2004;**41**:845–51.
- 11 McAndrews MP, Farcnik K, Carlen P, et al. Prevalence and significance of neurocognitive dysfunction in hepatitis C in the absence of correlated risk factors. *Hepatology* 2005;**41**:801–8.
- 12 Hilsabeck RC, Perry W, Hassanein TI. Neuropsychological impairment in patients with chronic hepatitis C. *Hepatology* 2002;**35**:440–6.
- 13 Meyerhoff DJ, Bloomer C, Cardenas V, et al. Elevated subcortical choline metabolites in cognitively and clinically asymptomatic HIV+ patients. *Neurology* 1999;**52**:995–1003.
- 14 Radkowski M, Wilkinson J, Nowicki M, et al. Search for hepatitis C virus negative-strand RNA sequences and analysis of viral sequences in the central nervous system: evidence of replication. *J Virol* 2002;**76**:600–8.
- 15 Forton DM, Karayiannis P, Mahmud N, et al. Identification of unique hepatitis C virus quasispecies in the central nervous system and comparative analysis of internal translational efficiency of brain, liver, and serum variants. *J Virol* 2004;**78**:5170–83.
- 16 Piche T, Vanbiervliet G, Cherikh F, et al. Effect of ondansetron, a 5-HT3 receptor antagonist, on fatigue in chronic hepatitis C: a randomised, double blind, placebo controlled study. *Gut* 2005;**54**:1169–73.
- 17 Capuron L, Neurauter G, Musselman DL, et al. Interferon-alpha-induced changes in tryptophan metabolism. Relationship to depression and paroxetine treatment. *Biol Psychiatry* 2003;**54**:906–14.
- 18 Kraus MR, Schaefer A, Faller H, et al. Paroxetine for the treatment of interferon-alpha-induced depression in chronic hepatitis C. *Aliment Pharmacol Ther* 2002;**16**:1091–9.
- 19 Wichers MC, Maes M. The psychoneuroimmunopathophysiology of cytokine-induced depression in humans. *Int J Neuropsychopharmacol* 2002;**5**:375–88.
- 20 Hayley S, Merali Z, Anisman H. Stress and cytokine-elicited neuroendocrine and neurotransmitter sensitization: implications for depressive illness. *Stress* 2003;**6**:19–32.
- 21 Bluthé RM, Michaud B, Kelley KW, et al. Vagotomy blocks behavioural effects of interleukin-1 injected via the intraperitoneal route but not via other systemic routes. *Neuroreport* 1996;**7**:2823–7.
- 22 Nguyen KT, Deak T, Owens SM, et al. Exposure to acute stress induces brain interleukin-1beta protein in the rat. *J Neurosci* 1998;**18**:2239–46.
- 23 Ramamoorthy S, Ramamoorthy JD, Prasad PD, et al. Regulation of the human serotonin transporter by interleukin-1 beta. *Biochem Biophys Res Commun* 1995;**216**:560–7.
- 24 Morikawa O, Sakai N, Obara H, et al. Effects of interferon-[alpha], interferon-[gamma] and cAMP on the transcriptional regulation of the serotonin transporter. *Eur J Pharmacol* 1998;**349**:317–24.
- 25 Kamata M, Higuchi H, Yoshimoto M, et al. Effect of single intracerebroventricular injection of [alpha]-interferon on monoamine concentrations in the rat brain. *Eur Neuropsychopharmacol* 2000;**10**:129–32.
- 26 Bonaccorso SM, Marino VM, Puzella AM, et al. Increased depressive ratings in patients with hepatitis C receiving interferon-[alpha]-based immunotherapy are related to interferon-[alpha]-induced changes in the serotonergic system. *J Clin Psychopharmacol* 2002;**22**:86–90.
- 27 Wichers MC, Maes M. The role of indoleamine 2,3-dioxygenase (IDO) in the pathophysiology of interferon-alpha-induced depression. *J Psychiatry Neurosci* 2004;**29**:11–17.
- 28 Schrocksnadel K, Wirleitner B, Winkler C, et al. Monitoring tryptophan metabolism in chronic immune activation. *Clin Chim Acta* 2006;**364**:82–90.
- 29 Fuchs D, Moller AA, Reibnegger G, et al. Increased endogenous interferon-gamma and neopterin correlate with increased degradation of tryptophan in human immunodeficiency virus type 1 infection. *Immunol Lett* 1991;**28**:207–11.
- 30 Fuchs D, Moller AA, Reibnegger G, et al. Decreased serum tryptophan in patients with HIV-1 infection correlates with increased serum neopterin and with neurologic/psychiatric symptoms. *J Acquir Immune Defic Syndr* 1990;**3**:873–6.

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