www.medscape.com

Chronic Opioid Use Is Associated With Altered Gut Microbiota and Predicts Readmissions in Patients With Cirrhosis

C. Acharya; N. S. Betrapally; P. M. Gillevet; R. K. Sterling; H. Akbarali; M. B. White; D. Ganapathy; A. Fagan; M. Sikaroodi; J. S. Bajaj

Aliment Pharmacol Ther. 2017;45(2):319-331.

Abstract and Introduction

Abstract

Background Opioid use is epidemic in cirrhosis, which could precipitate hepatic encephalopathy (HE) potentially through gut dysbiosis and inflammation.

Aim To define the effect of opioids on readmissions and on gut microbiota composition and functionality.

Methods Cohort 1 had 200 cirrhotic in-patients (with/without opioid use) followed prospectively through the index hospitalisation and 6 months post discharge. Readmissions (HE-related/unrelated) were compared between patients discharged on opioids compared to the rest, including using a multi-variable analysis. Cohort 2 consisted of 72 cirrhotics on chronic opioids who were age/model for end-stage liver disease (MELD) and prior HE-balanced with 72 cirrhotics not on opioids. Stool microbiota composition (multi-tagged sequencing), predicted functionality (PiCRUST), endotoxemia and systemic inflammation (IL-6, IL-17) were compared.

Results Cohort 1: Chronic opioid use was statistically similar between those admitted with/without HE, and was judged to be an HE precipitant in <5% of cases during the index hospitalisation. Of the 144 patients alive at 6 months, 82 were readmitted. The opioid users had a significantly higher all cause (69% vs. 48%, $P = 0.008$), but not HE-related readmissions (30% vs. 41%, $P = 0.30$). On regression, opioid therapy and female gender were predictive of readmission independent of MELD score and previous HE. Cohort 2: Significant dysbiosis was noted in the opioid cohort, especially in HE+opioid patients with lower autochthonous taxa and *Bacteroidaceae* relative abundance. PiCRUST showed highest aromatic amino acid and endotoxin production in opioid users. Opioid users also had higher endotoxemia and IL-6 but not IL-17.

Conclusion Chronic opioid use in cirrhosis is associated with increased endotoxemia, dysbiosis and all-cause readmissions.

Introduction

Prescription opioid use is epidemic in the USA and has been associated with a high morbidity and mortality.[1] Opioid use is particularly challenging in patients with chronic liver disease and cirrhosis, where chronic pain is prevalent and can be difficult to control.[2] This is partly because of the reluctance of clinicians to manage opioid doses due to the lower therapeutic-toxic threshold in cirrhotic patients and the relative contra-indications to concurrent acetaminophen and NSAIDs as alternatives.[3] The lower therapeutic-toxic threshold is partly due to reduced drug metabolism in the face of liver dysfunction and also the potential for precipitation of hepatic encephalopathy (HE) due to the associated constipation in opioid users.[4,5] Prior studies have also shown that opioid use in noncirrhotic models can directly lead to intestinal mucosal injury, along with microbial changes.[6,7] In out-patients with cirrhosis, opioid use has been associated with a higher healthcare utilisation focused on pain management.[2,8] However the impact on in-patients as well as the potential mechanisms behind these changes is unclear.

We hypothesised that opioid use on hospital discharge is associated with a higher risk of readmissions, specifically due to HE, and that cirrhotic patients with chronic opioid use have an unfavourable alteration in their gut microbiota composition and functionality.

Materials and Methods

We enrolled two different cirrhotic cohorts prospectively; cohort 1 consisted of cirrhotic patients who were hospitalised for non-elective reasons at VCU Medical Center while cohort 2 consisted of another set of out-patients with cirrhosis from VCU and McGuire VA Medical Centers. The study protocol was approved by the Institutional Review Boards at both medical centres.

Cohort 1

We enrolled patients with cirrhosis who were admitted for non-elective reasons who were able to give informed consent between April 2013 and December 2014. Cirrhosis was diagnosed using biopsy, Fibroscan, radiological evidence of cirrhosis, endoscopic evidence of varices in patients with chronic liver disease or frank decompensation. We excluded those who were admitted electively, with HIV infection or those with prior organ transplant. Patients were followed through this hospitalisation (termed index hospitalisation) till discharge and then for 6 months post discharge. Reasons for index admission, cirrhosis severity at admission and discharge, admission and discharge medications, and readmissions in the 6 months post-discharge period were collected. Opioid use on index admission, change (initiation, or discontinuation) during the hospitalisations, reason for opioid initiation, prescription of opioids on discharge and readmissions with/without HE in the context of chronic opioids was noted. We only analysed the first readmission after discharge from the index hospitalisation.

Cohort 2

A separate cohort of out-patients with cirrhosis was prospectively recruited from VCU Medical Center and McGuire VA Medical Center Hepatology clinics. All patients gave informed consent. Cirrhosis was diagnosed in similar methods as group 1 and we excluded subjects in whom cirrhosis was unclear, HIV infection, prior organ transplant, on antibiotics (including rifaximin), probiotics, recent alcohol use (within 3 months), or those who did not consent. We divided the group into those who were on and not on chronic opioid therapy (daily use for at least 3 months). We excluded rifaximin-using patients and patients on SBP prophylaxis given their impact on microbiota and because rifaximin use indicates a worse HE course. Patients on chronic opioid therapy were then model for end-stage liver disease (MELD) matched to an equal number of cirrhotic patients who were not on opioid therapy. All subjects underwent serum and stool collection on the same day as well as a 3-day food recall. Serum was analysed for endotoxin using LAL assay, and for IL-6 and IL-17 using ELISA at Assaygate, Ijamsville, MD using published techniques.[9] Stool was collected in RNA later with DNA extracted using published techniques.[10] We analysed the stool for microbiota composition using 16s sequencing.[11] Analysis of the microbiome was done for the entire group and separately for patients with and without prior HE using UNIFRAC, Kruskal–Wallis tests, Linear discriminant analysis effect size (LEFSe)[12] and predicted functionality using PiCRUST (phylogenetic investigation of communities using reconstruction of unobserved states).[13]

Statistical analysis was performed using *t*-test and chi-squared test as appropriate for patients who were or not on opioids during the index hospitalisation and readmission. Variables compared were demographics, cirrhosis severity, other medications, presence of HE and aetiology of cirrhosis. A multi-variable analysis with readmissions as the outcome was performed using backward logistic regression with variables that were significantly different between groups at least at $P < 0.10$, especially focusing on age, gender, MELD score, HE, infections, PPI use, SBP prophylaxis, hepatocellular cancer (HCC) and opioid use.

Results

In-patient Cohort 1

Between April 2013 and December 2014, we obtained informed consent from 200 patients during this index hospitalisation. Of the 200, 57 were admitted with opioids on board, in which opioid therapy was continued in 51 patients while six were withdrawn (four due to HE-related concern and two due to unclear reasons). Thirty-two cirrhotic patients were admitted without opioids, but were initiated on them during the hospitalisation (primarily for abdominal pain from ascites/hernias – Figure 1, and). The reasons for opioid use in these 51 and 32 patients are shown in .

Table 1. Description of the in-patient cohort

Variable	On opioids (*N* = 62)	Not on opioids (*N* = 82)	*P* value
Age (mean ± s.d.)	56.2 ± 9.7	55.8 ± 8.6	0.08
Male gender	37	48	0.89
Alcoholic aetiology	26	38	0.3
HCV aetiology	24	20	0.10
NASH aetiology	6	14	0.2
Other aetiologies	12	10	0.81
Hepatocellular cancer	10	6	0.10
Diabetes	26	26	0.2
Index hospitalisation details			
MELD score (mean ± s.d.)	20.2 ± 6.5	20.17 ± 5.3	0.98
Prior HE on admission	49	60	0.42
On lactulose only	20	30	0.59
On rifaximin also	29	30	0.22
On SBP prophylaxis	2	3	0.88
PPI use	24	38	0.68
HE during hospitalisation	20	36	0.15
Infection during hospitalisation	7	13	0.43
Readmission details			

All cause	43	39	0.008
HE	13	16	0.83
No-HE	30	23	0.012

We enrolled 200 patients during the initial admission, of which 144 were available for a 6-month post-discharge outcome assessment. Of these 144, 62 were opioids while 82 were not on opioids. The 62 'on opioids' patients shown here are patients discharged on opioids after the initial admission and the 82 'not on opioids' are patients who were not discharged on opioids after the initial admission. The patients on and not on opioids at 6 months post-discharge were similar on liver disease characteristics, complications during the index hospitalisation and demographics. However, the opioid-using patients had a higher rate of all cause and no-HE readmissions within 6 months of discharge.

Table 2. Reasons for narcotic use in the in-patient cohort

For patients admitted with narcotics on the index admission	**Count (*n* = 51)**	**For patients on whom narcotics were initiated on the index admission**	**Count (*n* = 32)**
GI related			
Pain from ascites-related abdominal distention	10	Pain from ascites-related abdominal distention	10
Abdominal hernia related pain	2	Abdominal hernia related pain	3
HCC-related abdominal pain	5	HCC-related abdominal pain	2
Chronic pancreatitis	3	Acute pancreatitis	2
SMV thrombosis	1	SMV thrombosis	1
Splenomegaly related abdominal pain	1		
Musculoskeletal/trauma			
Chronic extremity/back pain	19	Abdominal wall cellulitis	1
Extremity cellulitis/wound	3	Leg fracture	1
Gout	1	Gout flare	1
Septic arthritis	2	Septic arthritis	1
Fall and trauma	1	Leg fracture	1
		Post CPR pain	1
		Osteoarthritis related pain	1
Others			
CBD mass/cholangiocarcinoma	1	Post chest tube	1
Anal cancer	1	Dental infection	1
Breast cancer	1	Made hospice	5

In the in-patient cohort, 51 patients were admitted on opioids during the index admission, while 32 were initiated on them during the hospitalisation. The reasons for the opioid use at the time of admission and at the time of initiation during the hospitalisation are shown in the table and were not significantly different.

Table 2. Reasons for narcotic use in the in-patient cohort

For patients admitted with narcotics on the index admission	**Count (*n* = 51)**	**For patients on whom narcotics were initiated on the index admission**	**Count (*n* = 32)**
GI related			
Pain from ascites-related abdominal distention	10	Pain from ascites-related abdominal distention	10

Abdominal hernia related pain	2	Abdominal hernia related pain	3
HCC-related abdominal pain	5	HCC-related abdominal pain	2
Chronic pancreatitis	3	Acute pancreatitis	2
SMV thrombosis	1	SMV thrombosis	1
Splenomegaly related abdominal pain	1		
Musculoskeletal/trauma			
Chronic extremity/back pain	19	Abdominal wall cellulitis	1
Extremity cellulitis/wound	3	Leg fracture	1
Gout	1	Gout flare	1
Septic arthritis	2	Septic arthritis	1
Fall and trauma	1	Leg fracture	1
		Post CPR pain	1
		Osteoarthritis related pain	1
Others			
CBD mass/cholangiocarcinoma	1	Post chest tube	1
Anal cancer	1	Dental infection	1
Breast cancer	1	Made hospice	5

In the in-patient cohort, 51 patients were admitted on opioids during the index admission, while 32 were initiated on them during the hospitalisation. The reasons for the opioid use at the time of admission and at the time of initiation during the hospitalisation are shown in the table and were not significantly different.

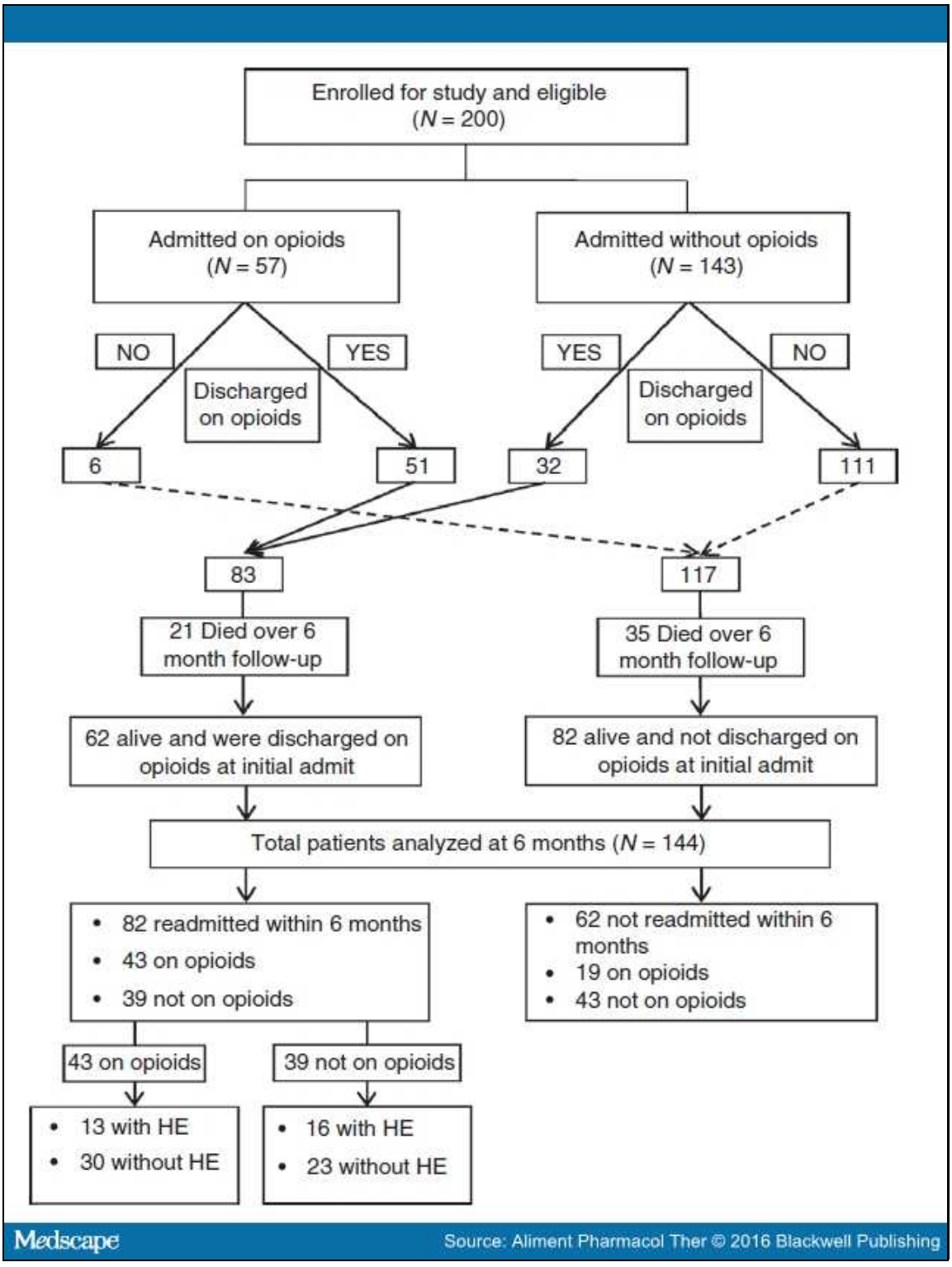

Figure 1.

Flow chart of the in-patient cohort (cohort 1) which follows each of the 200 patients initially consented during their index hospitalisation through changes in opioid use and 6 months, including readmissions.

Of the 200 patients, 79 developed or were hospitalised with HE during the index hospitalisation. The proportion of patients on chronic opioids as an out-patient before the index hospitalisation was statistically similar between those with and without HE on readmission (35.48% vs. 23.17%, *P* = 0.10). Of the patients who had HE during the index hospitalisation, chronic opioids were continued in 19, discontinued in four and initiated in nine. Six months post discharge, 56 of the 200 were not included in the analysis since they either died (*n* = 30) or were discharged to hospice (*n* = 26) at the end of the hospitalisation. Of these 56 excluded patients, 20 were on opioids (36%) compared to 62 (43%, *P* = 0.34) in those who survived the hospitalisation.

At the time of discharge from the index hospitalisation, there was no significant difference in the use of lactulose, rifaximin, PPI, SBP prophylaxis, cirrhosis aetiology, MELD score or proportion with HE in those on or not on opioids ( ).

Table 1. Description of the in-patient cohort

Variable	On opioids (*N* = 62)	Not on opioids (*N* = 82)	*P* value
Age (mean ± s.d.)	56.2 ± 9.7	55.8 ± 8.6	0.08
Male gender	37	48	0.89
Alcoholic aetiology	26	38	0.3
HCV aetiology	24	20	0.10
NASH aetiology	6	14	0.2
Other aetiologies	12	10	0.81
Hepatocellular cancer	10	6	0.10
Diabetes	26	26	0.2
Index hospitalisation details			
MELD score (mean ± s.d.)	20.2 ± 6.5	20.17 ± 5.3	0.98
Prior HE on admission	49	60	0.42
On lactulose only	20	30	0.59
On rifaximin also	29	30	0.22
On SBP prophylaxis	2	3	0.88
PPI use	24	38	0.68
HE during hospitalisation	20	36	0.15
Infection during hospitalisation	7	13	0.43
Readmission details			
All cause	43	39	0.008
HE	13	16	0.83
No-HE	30	23	0.012

We enrolled 200 patients during the initial admission, of which 144 were available for a 6-month post-discharge outcome assessment. Of these 144, 62 were opioids while 82 were not on opioids. The 62 'on opioids' patients shown here are patients discharged on opioids after the initial admission and the 82 'not on opioids' are patients who were not discharged on opioids after the initial admission. The patients on and not on opioids at 6 months post-discharge were similar on liver disease characteristics, complications during the index hospitalisation and demographics. However, the opioid-using patients had a higher rate of all cause and no-HE readmissions within 6 months of discharge.

Of the 200 patients, 52 were on opioids and discharged on them during the index admission. 31 were not on opioids and were initiated on opioids and discharged. Six were on opioids on the index admission and were taken off (five due to HE and one due to lack of indication). A total of 111 patients were not on opioids. Of the 144 patients included in the analysis for readmissions, 62 were on opioids (52 were already on these as out-patients and 10 were initiated during this hospitalisations) and 82 were not on opioids. Of the 82 not sent home on opioids post-index admission, four had been admitted with opioids and that was stopped during the index admission (all for HE precipitating concerns at the time of discharge). The specific opioids used were hydromorphone (*n* = 7), fentanyl (1), methadone (1), morphine sulphate (1), oxycodone (23), Percocet, (3) tramadol

(23) and combinations of the drugs (3). The patients were followed up for a median of 25 days (IQR 10–67 days) during which 82 were readmitted. None of the patients were seen in the ER or clinics to increase opioid dosage once discharged. The leading causes of readmission have been listed in and shows HE as the leading cause in both subgroups. Of the 62 patients on opioids, 43 were readmitted and of the 82 not on opioids 39 were readmitted. Interestingly, non-HE admissions were higher in the patients on opioids compared to those without opioids while HE-related admissions were statistically similar ( ). No particular therapy for HE was predominant have factored into readmission. On evaluation of the type of opioid prescribed for the group readmitted with HE, oxycodone (*n* = 8) was noted to be the first choice of opioid, while Percocet (1), tramadol (2), Morphine sulphate (1) and hydromorphone (1) were less common (not shown in tables).

Table 3. Readmission aetiologies

Readmission aetiology for pts discharged on opioids	Count (*N* = 43)	Readmission aetiology for pts not discharged on opioids	Count (*N* = 39)
GI-related			
Hepatic encephalopathy	13	Hepatic encephalopathy	16
Ascites/volume overload/AKI	7	Ascites/volume overload/AKI	5
GI bleed	2	GI bleed	1
Nausea/vomiting	4	Nausea/vomiting	0
Infections			
SBP	2	SBP	1
Pneumonia	2	Pneumonia	1
Cellulitis	1	Diverticulitis	1
UTI	2		
Transplant related			
Liver transplant	2	Liver transplant	2
Other causes and procedures			
Dental extraction	1	Dental extraction	1
Hernia repair	2	Hernia repair	3
Syncope/fall	2	Syncope/Fall	2
Post-tips bleeding	1	Chest pain	1
Hematoma related pain	1	Chronic pancreatitis	1
		Hypoglycaemia	1
		CHF exacerbation	1
		Alcohol-related	2

Of the 144 patients analysed in the in-patient cohort at 6 months, 62 were on opioids and 82 were not on opioids. Of the 62 patients not on opioids, 43 were readmitted in 6 months, while in the 82 not on opioids at 6 months, 39 patients were readmitted. Hepatic encephalopathy-unrelated related readmissions total were significantly higher in the opioid group while hepatic encephalopathy-related readmissions were similar. This was primarily driven by the non-HE GI related causes (*P* = 0.03). No others subgroup differences (infections, other causes, etc.) were identified.

Table 1. Description of the in-patient cohort

Variable	On opioids (*N* = 62)	Not on opioids (*N* = 82)	*P* value
Age (mean ± s.d.)	56.2 ± 9.7	55.8 ± 8.6	0.08
Male gender	37	48	0.89

Alcoholic aetiology	26	38	0.3
HCV aetiology	24	20	0.10
NASH aetiology	6	14	0.2
Other aetiologies	12	10	0.81
Hepatocellular cancer	10	6	0.10
Diabetes	26	26	0.2
Index hospitalisation details			
MELD score (mean ± s.d.)	20.2 ± 6.5	20.17 ± 5.3	0.98
Prior HE on admission	49	60	0.42
On lactulose only	20	30	0.59
On rifaximin also	29	30	0.22
On SBP prophylaxis	2	3	0.88
PPI use	24	38	0.68
HE during hospitalisation	20	36	0.15
Infection during hospitalisation	7	13	0.43
Readmission details			
All cause	43	39	0.008
HE	13	16	0.83
No-HE	30	23	0.012

We enrolled 200 patients during the initial admission, of which 144 were available for a 6-month post-discharge outcome assessment. Of these 144, 62 were opioids while 82 were not on opioids. The 62 'on opioids' patients shown here are patients discharged on opioids after the initial admission and the 82 'not on opioids' are patients who were not discharged on opioids after the initial admission. The patients on and not on opioids at 6 months post-discharge were similar on liver disease characteristics, complications during the index hospitalisation and demographics. However, the opioid-using patients had a higher rate of all cause and no-HE readmissions within 6 months of discharge.

On logistic regression, the only significant variables predictive of readmissions were opioid use (P = 0.006, OR = 2.7, CI = 1.3–6.1) and female gender (P = 0.02, OR = 2.44, CI = 1.14–5.39). This was independent of MELD score, HE (lactulose or rifaximin use), cirrhosis aetiology, infections, HCC, PPI use and SBP prophylaxis and age.

Out-patient Cohort Group 2

In this cohort, there were 72 cirrhotic out-patients who were on chronic opioids. These were age and MELD-matched to 72 cirrhotic patients who were not on any opioid therapy. There was a statistically similar number of VAMC and VCU patients in opioid users or non-users (25 VA opioid users and 20 VA opioid non-users) with similar aetiology distributions (HCV VA 29 of 45 total, 14 users and 15 non-users, Alcohol VA 11 total, six users and five non-users and NASH 5 total two users, three non-users). There were no significant differences with respect to comorbid conditions, cirrhosis severity or aetiology of cirrhosis. Opioid-using patients were on therapy for a median of 5 months (IQR 3–15 months) of which seven were on tramadol, 42 on oxycodone, 11 on morphine, eight on hydromorphone and four on methadone. The reasons for the opioid initiation were related to chronic back pain (n = 29), chronic musculoskeletal pain (n = 18), abdominal pain (n = 13), HCC-related (n = 6) and others (n = 10). Both users and non-users were equivalent on cirrhosis severity and all HE patients were only on lactulose ( ).

Table 4. Description of the out-patient cohort that underwent microbiota analysis

Variable	On opioids (N = 72)	Not on opioids (N = 72)	P value
Age	56.8 ± 6.1	57.2 ± 6.7	0.32
Male gender (%)	60 (83)	62 (86)	0.89
BMI	29.6 ± 6.4	29.3 ± 5.0	0.62

24 h caloric intake	2014 ± 290	2879 ± 329	0.42
HCV aetiology	43	41	0.24
Alcoholic aetiology	20	13	0.17
NASH aetiology	7	11	0.31
Other aetiologies	2	7	0.09
Diabetes (%)	20 (28)	21 (29)	0.83
MELD score	15.9 ± 7.4	15.9 ± 6.6	0.51
Daily bms (HE pts median IQR)	1 (0–2)	1 (1–3)	0.24
Daily bms (no-HE pts median IQR)	2 (1–4)	2 (2–5)	0.34
HE, *n* (%)	40 (56)	38 (53)	0.74
Proton pump inhibitor use, *n* (%)	42 (58)	37 (51)	0.85

This table demonstrates the demographics, cirrhosis characteristics and medication use of the out-patient cohort in which 72 cirrhotic out-patients who were chronic opioid users were age and MELD-matched to 72 cirrhotic out-patients who were not on opioids. No significant differences on any characteristics were demonstrated.

There was a significant increase in endotoxin [median (IQR) 0.09 (0.47) vs. 0.02 (0.13) EU/mL, $P = 0.03$] and IL-6 [49.0 (7.9) vs. 3.43 (12.5) pg/mL, $P = 0.028$] vs. in opioid users compared to those not on opioids. IL-17 levels were similar between groups [0.95 (7.2) vs. 0.90 (2.9) pg/mL, $P = 0.84$].

On individual analyses for the entire group using Kruskal–Wallis test, there was a significantly reduced median relative abundance of the autochthonous families in cirrhotics on opioids compared to others (Clostridiales XIV and *Lachnospiraceae*) along with a decrease in *Bacteroidaceae* relative abundance (Figure 2).

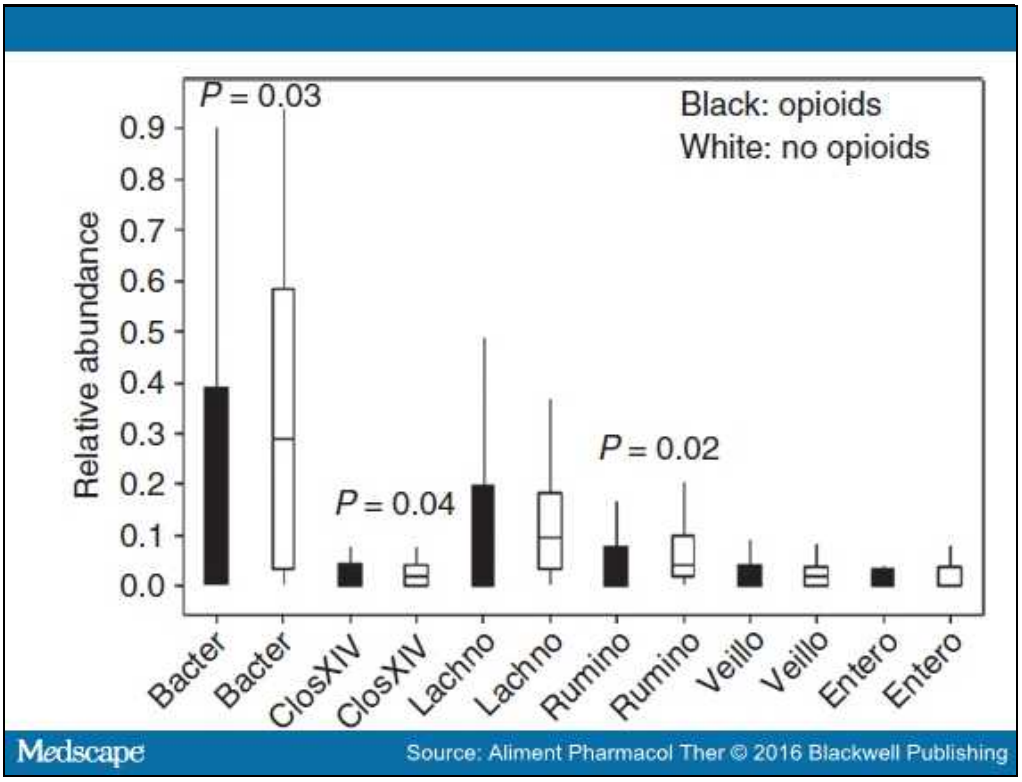

Figure 2.

In the out-patient cohort (cohort 2) the Kruskal–Wallis of the relative abundances of gut microbiota in patients with/without opioid use are demonstrated. Median and IQR of

relative abundances are shown and significant differences are shown; black bars indicate opioid users and white bars indicate non-opioid users. Specific families shown here are *Bacter: Bacteroidaceaeae, Lachno: Lachnospiraceae, Rumino: Ruminococcaceae, Clos XIV: Clostridiales XIV, Veillo: Veillonellaceae* and *Entero: Enterobacteriacae*. There were significant reductions in the relative abundances of authochthonous or beneficial taxa (*Ruminococcaceae* and Clostridiales XIV) in the opioid users in the out-patient cohort compared to non-users.

UNIFRAC was significantly different between HE patients on opioids compared to HE patients not on opioids ($P = 0.05$) as well as in no-HE patients on opioids vs. no-HE patients not on opioids ($P = 0.03$). On LEFSe at the genus level, there was a significant change in microbiota between patients on opioids with HE compared to HE patients not on opioids as well as in the no-HE group (Figure 3a,b). Autochthonous taxa were significantly lower in HE on opioids while *Bifidobacterium* was higher. In no-HE patients, microbiota differences between patients on or not on opioids were relatively smaller with decreased *Parasutterella* and higher *Peptostreptococcaceae* in the opioid users (Figure 3c,d).

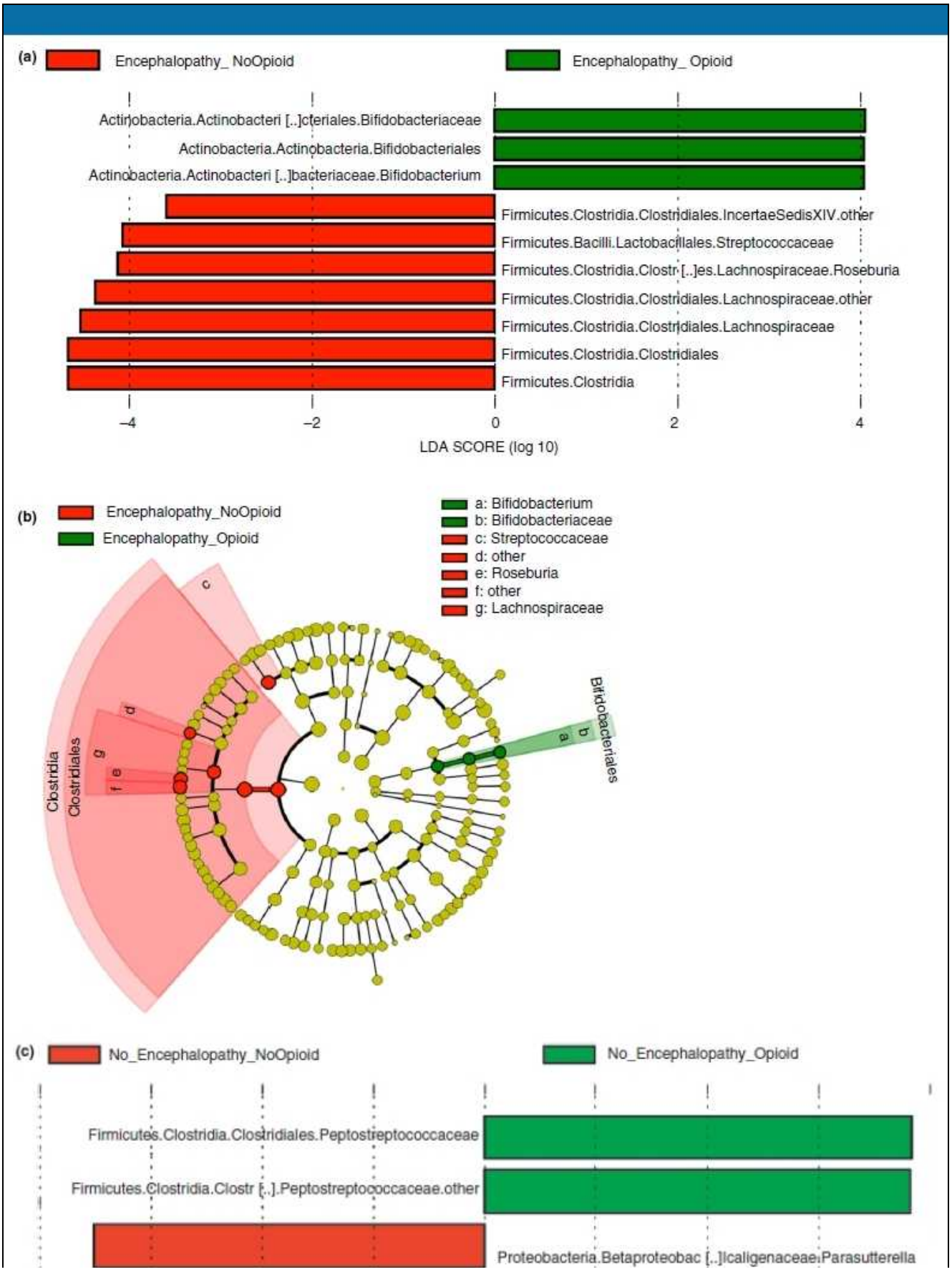
(a)
Encephalopathy_ NoOpioid
Encephalopathy_ Opioid
Actinobacteria.Actinobacteri [..]cteriales.Bifidobacteriaceae
Actinobacteria.Actinobacteria.Bifidobacteriales
Actinobacteria.Actinobacteri [..]bacteriaceae.Bifidobacterium
Firmicutes.Clostridia.Clostridiales.IncertaeSedisXIV.other
Firmicutes.Bacilli.Lactobacillales.Streptococcaceae
Firmicutes.Clostridia.Clostr [..]es.Lachnospiraceae.Roseburia
Firmicutes.Clostridia.Clostridiales.Lachnospiraceae.other
Firmicutes.Clostridia.Clostridiales.Lachnospiraceae
Firmicutes.Clostridia.Clostridiales
Firmicutes.Clostridia
−4
−2
0
2
4
LDA SCORE (log 10)
(b)
Encephalopathy_NoOpioid
Encephalopathy_Opioid
a: Bifidobacterium
b: Bifidobacteriaceae
c: Streptococcaceae
d: other
e: Roseburia
f: other
g: Lachnospiraceae
c
d
g
f e
Clostridia
Clostridiales
a
b
Bifidobacteriales
(c)
No_Encephalopathy_NoOpioid
No_Encephalopathy_Opioid
Firmicutes.Clostridia.Clostridiales.Peptostreptococcaceae
Firmicutes.Clostridia.Clostr [..].Peptostreptococcaceae.other
Proteobacteria.Betaproteobac [..]lcaligenaceae.Parasutterella

Figure 3.

In the out-patient cohort, we separately compared microbiota differences between patients with and without HE and opioid use. (a, b) In all out-patients with HE, we compared those with HE on opioids to those with HE who were not on opioids using LEFSe. There were significant differences using histogram and cladogram; red bars indicate patients with HE without opioids while green bars indicate patients with HE on opioids. As expected, autochthonous taxa (*Lachnospiraceae*, Clostridiales XIV) were significantly higher in the HE group not on opioids. (c, d) In all out-patients without HE, we compared those without HE on opioids to those without HE not on opioids on LEFSe. There shows significant differences using histogram and cladogram; red bars indicate patients without HE without opioids while green bars indicate patients without HE on opioids. The differences were relatively minor compared to those in the HE group.

When the specific aetiologies were compared, NASH aetiology was associated with greater dysbiosis in patients without opioids but this difference disappeared in opioid users (Table S1). Alcoholic aetiology did not impact between the microbial dysbiosis in this selected population.

Using PiCRUST in patients with opioids as a whole, metabolism of aromatic amino acids and degradation of potentially beneficial branched-chain amino acids were noted in opioid-associated microbiota (Figure 4a). In contrast, branched-chain amino acid synthesis and bio-energetics processes were over-represented in non-opioid users. In HE patients on opioids, the gut microbiota were significantly predictive for the endotoxin and endotoxin protein synthesis, nitrogen metabolism, motility and degradation of the branched-chain amino acids compared to those with HE not on opioids (Figure 4b). In opioid-using no-HE patients, the predicted bacterial functionality was related to aromatic amino acids compared to the no-HE opioid users (Figure 4c).

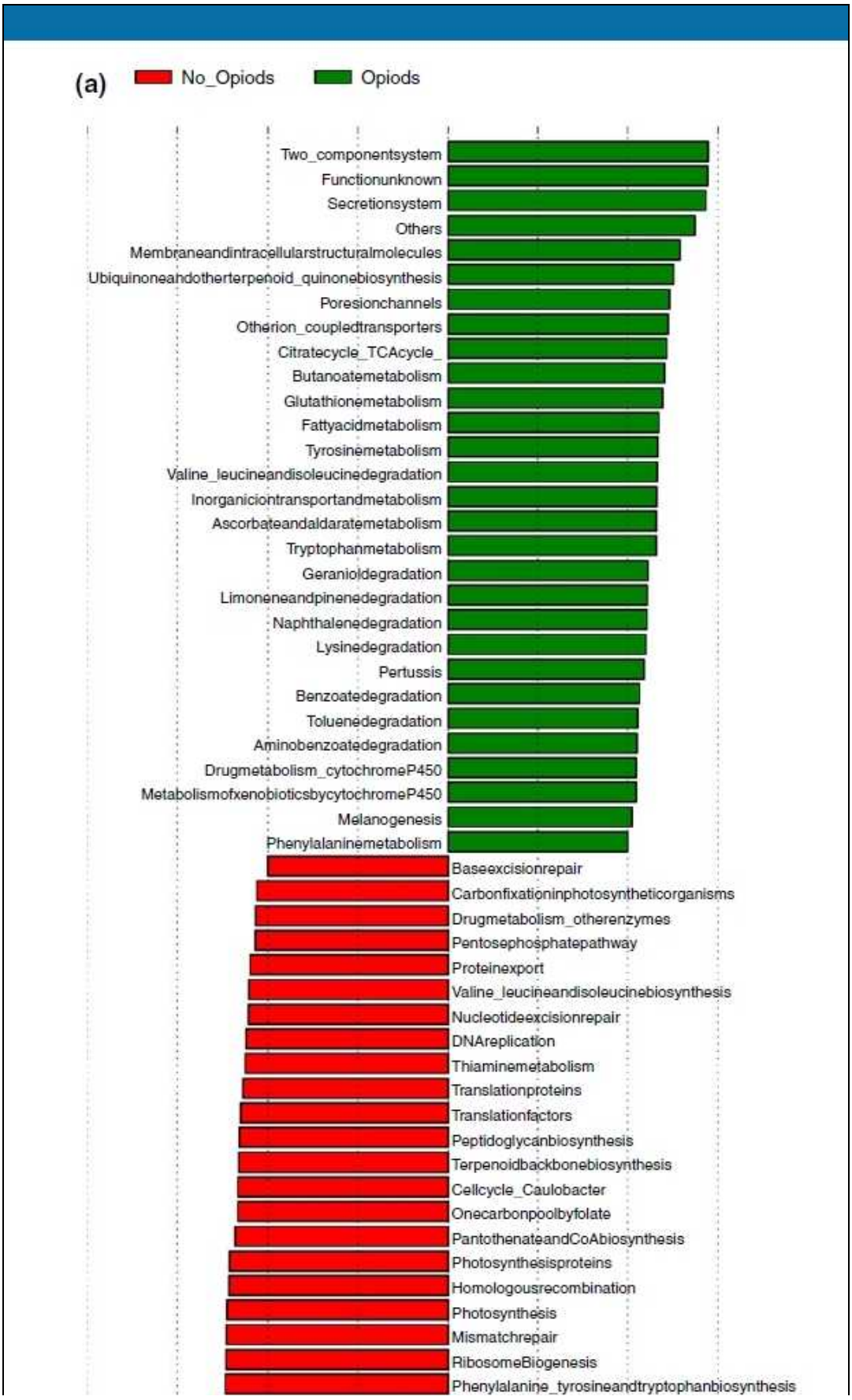
(a)
No_Opiods
Opiods
Two_componentsystem
Functionunknown
Secretionsystem
Others
Membraneandintracellularstructuralmolecules
Ubiquinoneandotherterpenoid_quinonebiosynthesis
Poresionchannels
Otherion_coupledtransporters
Citratecycle_TCAcycle_
Butanoatemetabolism
Glutathionemetabolism
Fattyacidmetabolism
Tyrosinemetabolism
Valine_leucineandisoleucinedegradation
Inorganiciontransportandmetabolism
Ascorbateandaldaratemetabolism
Tryptophanmetabolism
Geranioldegradation
Limoneneandpinenedegradation
Naphthalenedegradation
Lysinedegradation
Pertussis
Benzoatedegradation
Toluenedegradation
Aminobenzoatedegradation
Drugmetabolism_cytochromeP450
MetabolismofxenobioticsbycytochromeP450
Melanogenesis
Phenylalaninemetabolism
Baseexcisionrepair
Carbonfixationinphotosyntheticorganisms
Drugmetabolism_otherenzymes
Pentosephosphatepathway
Proteinexport
Valine_leucineandisoleucinebiosynthesis
Nucleotideexcisionrepair
DNAreplication
Thiaminemetabolism
Translationproteins
Translationfactors
Peptidoglycanbiosynthesis
Terpenoidbackbonebiosynthesis
Cellcycle_Caulobacter
Onecarbonpoolbyfolate
PantothenateandCoAbiosynthesis
Photosynthesisproteins
Homologousrecombination
Photosynthesis
Mismatchrepair
RibosomeBiogenesis
Phenylalanine_tyrosineandtryptophanbiosynthesis

Figure 4.

The predicted functionality of the gut microbiota was studied using PiCRUST for the entire cohort and then separately between patients with and without HE and opioid use. Data are presented in a histogram with Linear Discriminant Analysis between groups. (a) Between opioid users and non-users in the entire out-patient cohort: Green bars indicate predicted function in gut microbiota of patients on opioids while red bars indicate the same for patients not on opioids. Predicted functions related to branched amino acid degradation and aromatic amino acid metabolism were significantly higher in the microbiota in opioid using cirrhotic out-patients. (b) Between HE patients in the out-patient cohort on and not on opioids: PiCRUST showing predicted bacterial functionality more in patients with HE on opioids (green bars) compared to patients with HE not on opioids (red bars). Predicted functions related to endotoxin synthesis, branched amino acid degradation and aromatic amino acid metabolism were significantly higher in the microbiota in opioid-using HE cirrhotic out-patients. (c) Between patients without HE in the out-patient cohort on and not on opioids: PiCRUST showing predicted bacterial functionality more in patients without HE on opioids (green bars) compared to patients without HE not on opioids (red bars). Predicted functions related to aromatic amino acid metabolism were significantly higher in the microbiota in opioid-using cirrhotic out-patients without HE compared to those who were not on opioids.

Discussion

This study results demonstrate that chronic opioid therapy is associated with a higher risk of readmissions in cirrhotic patients independent of HE or other currently available biomarkers. This could be partly explained by worsening dysbiosis, endotoxemia and systemic inflammatory milieu in cirrhotic patients who are on opioids compared to others despite matching for liver disease severity.

Opioid misuse remains a public health emergency and its negative impact may be magnified in patients with chronic diseases such as cirrhosis. Opioid use in advanced liver disease patients is particularly fraught since many opioids are metabolised in the liver, which could affect their drug metabolism.[3] The use is also restricted in this population due to the potential for HE causation due to constipation or alteration in mental status. There are various proposed mechanisms for impaired drug metabolism, such as hypoalbuminemia-associated increase in drug availability and reduction in the number of functional hepatocytes.[5] Animal studies have also examined the role of *μ* opioid receptors as a possible potentiators of HE.[14] Opioid receptor ligands have also noted to be elevated in HE patients.[15] However, a systematic clinical evaluation of opioid-associated effect on cirrhosis outcomes is unclear.

In the in-patient study, despite a significant proportion of cirrhotics being on opioids for their HE admission, a minority was adjudged to be solely precipitated due to the opioids. This could be due to the chronicity of use which could have decreased the clinician's focus on these medications or potentially a synergistic role along with the predominant precipitating factor in the ultimate precipitation of the overt HE episode. Most opioids were continued during the hospitalisation and even initiated during the hospitalisation, regardless of in-patient HE development. As shown in the results, these were due to pain issues related to abdominal pain, musculoskeletal issues and in some cases due to terminal pain concerns. The initiation of these medications could also indicate that in the assessment of the prescribing physician, the risk/benefit ratio was skewed towards treating pain that was acute and distressing rather than towards dealing with a potential future risk of HE in these patients. Interestingly, opioid therapy was continued in most patients who were admitted with it on board. Most chronic opioid use in patients admitted with opioids in the in-patient cohort and in the out-patient cohort was related to chronic musculoskeletal and predominantly lower back pain. It is likely that opioids were continued in patients in whom they were considered noncontributory to the index admission. However, patients in the in-patient cohort who had their opioid therapy continued or initiated due to ascites-related abdominal pain could potentially be ameliorated by large volume paracenteses and judicious management of ascites alone. The numerous aetiologies for opioid initiation and continuation in advanced cirrhosis reiterate the complexity of managing chronic pain in end-stage liver disease.[8] This could justify a dedicated in-patient multi-modal care plan for pain management with an adequate transition phase when the patient is discharged.

The shadow of opioid use extends beyond the index hospitalisation with a significantly higher risk of all-cause readmission within 6 months of hospital discharge, which was independent of MELD score and HE. However, opioid use was not specifically associated with HE readmissions but only for non-HE reasons. This raises important concerns about the role of opioids in worsening clinically relevant outcomes beyond that are usually expected by treating clinicians. This dichotomy could be due to a relatively lower sample size once the subdivisions were performed or could be due to a greater awareness of the association of HE with opioids, but not an association of opioids and all-cause readmissions. These results are consistent with Tapper *et al*., who demonstrated that substance abuse also can independently impact readmission.[16] Given the relatively advanced cirrhosis in our in-patient cohort, the readmission rate is higher but we also extended the period to 6-months rather than the usual 30 and 90-day published results.[16,17] Since the opioids were continued in the majority of the in-patient cohort that was on opioids, it is also clear that that none of the patients sought clinical attention for dose increases or were indeed seen in the ER for chronic pain in between these hospitalisations. This represents a difficult balancing act for clinicians to undertake given the competing risks of readmissions for medical conditions vs. medical attention for pain-related issues as has been shown before.[2]

Although it is reasonable to assume that opioids may result in constipation causing small bowel overgrowth and resultant bacterial translocation, the impact of opioids is indeed multi-factorial. In animal models of sepsis, opioid use significantly worsens the course through modulation of IL-17 that is associated with Gram-positive organism infections.[7] Other animal studies have shown that opioids increase the virulence of Gram-negative organisms as well such as *Pseudomonas aeruginosa*.[18] There is also evidence of direct mucosal barrier disruption in animals exposed to opioids, even using the subcutaneous, nongut route.[6] The human data demonstrate a significantly higher pro-inflammatory milieu characterised by endotoxemia and IL-6, but not IL-17 increase in opioid using out-patient cirrhotics compared to age and MELD-matched non-opioid users.[19] The lack of IL-17 change, which is often associated with Gram-positive sepsis, is likely due to predominant Gram-negative relative abundance usually observed in

cirrhosis.[7] Indeed most cirrhosis-associated infections and prophylaxis strategies are geared towards Gram-negative organisms that produce endotoxin.[20] This was accompanied by a dysbiosis characterised by reduction in autochthonous bacteria and *Bacteroidaceae*. *Bacteroidaceae* abundance prevents the overgrowth of potentially harmful nosocomial pathogens and their relative reduction has been shown to promote hospitalisations; therefore, the complicity of opioids in doing this may be a factor towards this.[21] Interestingly, the worse dysbiosis seen in NASH cirrhotic patients without opioids in the out-patient cohort disappeared in opioid users, indicating specific modulation of the microbiota by disease aetiology that can be further affected by superadded opioid use.[22]

Autochthonous taxa, which were reduced in the out-patient opioid using cohort, are considered markers of gut health across several studies and have important functions related to short chain fatty acid and bile acid metabolism.[23] This is similar to a recent noncirrhotic animal study in which opioid use reduced bile acid profile suggestive of autochthonous taxa reduction.[6] In addition to the change in bacterial composition, the predicted functionality pointed towards a higher endotoxin and endotoxin binding protein synthesis, and aromatic amino acid metabolic function in gut microbiota of opioid-using cirrhotic patients.[9] These striking findings help frame the discussion of opioid use in cirrhosis by controlling for most confounders such as aetiology of cirrhosis,[24,25] cirrhosis severity, use of lactulose, PPI use and daily bowel movement frequency, all of which were similar between the matched groups. This dysbiosis, with altered predicted microbial functionality, likely transcends the relatively simple concept of slow intestinal transit. This could form the basis of a higher systemic inflammatory milieu that predicts worsening outcomes as shown in the in-patient study.

Our study is limited by the use of two separate patient cohorts, which was performed to avoid the multiple concomitant confounders of the in-patient microbiota analysis. The reasons for chronic opioid use were similar across both groups, which can increase the generalisability of the results. We did not find a high pain-related medical behavior but rather readmissions for GI-related issues after discharge in the in-patient cohort, which may be due to the more advanced nature of their disease process compared to prior studies. It may also be likely that opioids would have been withheld or not prescribed in the most advanced cirrhotic in-patients, therefore, further modulating the HE-related readmission risk in these patients. It is possible that other factors could predict readmissions and changes in microbiota; however, multi-variable adjustments and matching were performed based on known variables that can influence readmissions and microbiota composition and functionality. We excluded rifaximin-using patients from the out-patient cohort to account for its impact on microbiota function and to aid in matching by MELD score.[26] This is because patients on rifaximin are more advanced than those just on lactulose due to the US prescribing practices.[27] Despite excluding the group with the highest potential dysbiosis (advanced cirrhosis on rifaximin), we were still able to demonstrate differences in microbiota. We used both VAMC and VCU patients in the out-patient but only VCU patients in the in-patient cohort. However, the demographics, aetiology distribution and cirrhosis-related characteristics were similar between groups. Due to the small subgroup size, all opioids were considered equivalent in promoting these negative outcomes and future studies are required for study separate opioids. However, only a minority of patients was on tramadol, which is considered relatively safe in cirrhosis.

We conclude that opioid use, initiation and continuation, are prevalent in cirrhosis, which may be associated with all cause but not HE-related readmissions. Alterations in gut microbiota composition and functionality, and systemic inflammation are associated with opioid use in cirrhotic patients, which occurs in patients with cirrhosis regardless of prior HE status, could be a potential contributing factor. Cirrhotic patients with chronic opioid use are a potential high-risk group that may benefit from close monitoring using a multi-modal pain management team that has the capability of transitioning between in-patient and out-patient settings.

References

1. Website at Department of Health and Human Services http://www.hhs.gov/sites/default/files/Factsheet-opioids-061516.pdf. Accessed 1 August 2016.
2. Rogal SS, Winger D, Bielefeldt K, Szigethy E. Pain and opioid use in chronic liver disease. *Dig Dis Sci* 2013; 58: 2976–85.
3. Verbeeck RK. Pharmacokinetics and dosage adjustment in patients with hepatic dysfunction. *Eur J Clin Pharmacol* 2008; 64: 1147–61.
4. Vilstrup H, Amodio P, Bajaj J, *et al.* Hepatic encephalopathy in chronic liver disease: 2014 Practice Guideline by the American Association for the Study of Liver Diseases and the European Association for the Study of the Liver. *Hepatology* 2014; 60: 715–35.
5. Lewis JH, Stine JG. Review article: prescribing medications in patients with cirrhosis – a practical guide. *Aliment Pharmacol Ther* 2013; 37: 1132–56.
6. Banerjee S, Sindberg G, Wang F, *et al.* Opioid-induced gut microbial disruption and bile dysregulation leads to gut barrier compromise and sustained systemic inflammation. *Mucosal Immunol* 2016; 9: 1418–28
7. Meng J, Banerjee S, Li D, *et al.* Opioid exacerbation of Gram-positive sepsis, induced by gut microbial modulation, is rescued by IL-17A neutralization. *Sci Rep* 2015; 5: 10918.
8. Rogal SS, Bielefeldt K, Wasan AD, *et al.* Inflammation, psychiatric symptoms, and opioid use are associated with pain and disability in patients with cirrhosis. *Clin Gastroenterol Hepatol* 2015; 13: 1009–16.
9. Bajaj JS, Heuman DM, Hylemon PB, *et al.* Altered profile of human gut microbiome is associated with cirrhosis and its complications. *J Hepatol* 2014; 60: 940–7.
10. Bajaj JS, Betrapally NS, Gillevet PM Decompensated cirrhosis and microbiome interpretation. *Nature* 2015; 525: E1–2.

11. Gillevet P, Sikaroodi M, Keshavarzian A, Mutlu EA. Quantitative assessment of the human gut microbiome using multitag pyrosequencing. *Chem Biodivers* 2010; 7: 1065–75.

12. Segata N, Izard J, Waldron L, *et al.* Metagenomic biomarker discovery and explanation. *Genome Biol* 2011; 12: R60.

13. Langille MG, Zaneveld J, Caporaso JG, *et al.* Predictive functional profiling of microbial communities using 16S rRNA marker gene sequences. *Nat Biotechnol* 2013; 31: 814–21.

14. Bergasa NV, Rothman RB, Mukerjee E, Vergalla J, Jones EA. Up-regulation of central mu-opioid receptors in a model of hepatic encephalopathy: a potential mechanism for increased sensitivity to morphine in liver failure. *Life Sci* 2002; 70: 1701–8.

15. Yurdaydin C, Karavelioglu D, Onaran O, Celik T, Yasa MH, Uzunalimoglu O. Opioid receptor ligands in human hepatic encephalopathy. *J Hepatol* 1998; 29: 796–801.

16. Tapper EB, Halbert B, Mellinger J. Rates of and reasons for hospital readmissions in patients with cirrhosis: a multistate population-based cohort study. *Clin Gastroenterol Hepatol* 2016; 14: 1181–8.e2.

17. Kanwal F, Asch SM, Kramer JR, Cao Y, Asrani S, El-Serag HB. Early outpatient follow-up and 30-day outcomes in patients hospitalized with cirrhosis. *Hepatology* 2016; 64: 569–81.

18. Babrowski T, Holbrook C, Moss J, *et al. Pseudomonas aeruginosa* virulence expression is directly activated by morphine and is capable of causing lethal gut-derived sepsis in mice during chronic morphine administration. *Ann Surg* 2012; 255: 386–93.

19. Shawcross DL. Is it time to target gut dysbiosis and immune dysfunction in the therapy of hepatic encephalopathy? *Expert Rev Gastroenterol Hepatol* 2015; 9: 539–42.

20. Tandon P, Garcia-Tsao G. Bacterial infections, sepsis, and multiorgan failure in cirrhosis. *Semin Liver Dis* 2008; 28: 26–42.

21. Bajaj JS, Betrapally NS, Hylemon PB, *et al.* Gut microbiota alterations can predict hospitalizations in cirrhosis independent of diabetes mellitus. *Sci Rep* 2015; 5: 18559.

22. Boursier J, Mueller O, Barret M, *et al.* The severity of nonalcoholic fatty liver disease is associated with gut dysbiosis and shift in the metabolic function of the gut microbiota. *Hepatology* 2016; 63: 764–75.

23. Nava GM, Stappenbeck TS. Diversity of the autochthonous colonic microbiota. *Gut Microbes* 2011; 2: 99–104.

24. Wieland A, Frank DN, Harnke B, Bambha K. Systematic review: microbial dysbiosis and nonalcoholic fatty liver disease. *Aliment Pharmacol Ther* 2015; 42: 1051–63.

25. Vassallo G, Mirijello A, Ferrulli A, *et al.* Review article: alcohol and gut microbiota – the possible role of gut microbiota modulation in the treatment of alcoholic liver disease. *Aliment Pharmacol Ther* 2015; 41: 917–27.

26. DuPont HL. Review article: the antimicrobial effects of rifaximin on the gut microbiota. *Aliment Pharmacol Ther* 2016; 43(Suppl 1): 3–10.

27. Website describing rifaximin prescribing information at the FDA. http://www.accessdata.fda.gov/drugsatfda_docs/label/2010/022554lbl.pdf. Accessed 1 August 2016.

Authorship
Guarantor of the article
Jasmohan Bajaj.

Aliment Pharmacol Ther. 2017;45(2):319-331.