with 8 incidents relating to ongoing NGT position testing. 2 NGTs moved without a change in nostril measurement; both identified via X-ray, not pH testing. 1 tube was in the lung and caused low harm. The other was in the oesophagus caused no harm. There were another 2 recorded incidents of missed medications and/or feed due to failed ongoing pH tests.

Conclusions These results highlight that NGTs can spontaneously displace and pH testing does not always identify these. It also indicates that failed pH test results can and do lead to delays in feeding and medications. Incident reporting likely captures only a fraction of these adverse outcomes and further primary observational research is required for more accurate representation.

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Colon and anorectum

PTH-90 PREVALENCE OF CLOSTRIDIOIDES DIFFICILE INFECTION IN CENTRAL INDIA: A PROSPECTIVE OBSERVATIONAL COHORT STUDY

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Introduction The true burden of *Clostridioides difficile* infection (CDI) in India remains poorly understood. Prolifigate, unregulated antibiotic use and inappropriate prescribing suggest that CDI could be widespread in India. Our aim was to establish and compare baselines rates of CDI in both in-and outpatient settings in Nagpur city district and rural Melghat, Central India.

Methods We recruited adult participants aged ≥18 years of age who could provide written or thumb-print informed consent. A diagnosis of diarrhoea was defined as 3 or more loose stools in a 24-hour period. Immunosuppression was defined as those on prednisolone (>5mg/day), immunomodulators or biologics. Baseline characteristics were also collected and included: demographics, symptomatology, antibiotics exposure, duration of diarrhoea, hospitalisation status at recruitment, and duration, BMI, animal exposure, housing conditions, toilet access, and seasonality. All diarrhoeal samples were tested for CDI using the C. DIFF QUIK CHEK COMPLETE-enzyme immunoassay in accordance with the manufacturers' instructions.

Results *C. difficile* testing was performed on 1223 patients with acute diarrhoea. A total of 36 patients (2.9%) tested positive for both GDH antigen and toxin expression. A higher% of urban inpatient diarrheal samples tested positive for toxigenic *C. difficile* (26 cases; 8%) compared to that seen for urban outpatients (9 cases; 3%) and the rural diarrhoeal group (1 outpatient case). Of those testing positive for toxigenic *C. difficile*, 63.9% were immunosuppressed and almost all (94.4%) were on antibiotics at the time of recruitment. The majority of the toxigenic CDI cases were detected during the monsoon season, lived in very good or good housing conditions, had access to good toilet facilities and reported no cohabitation with animals. Non-toxigenic *C. difficile* was detected in 6.2%, 4.8%, and 0.5% in the urban inpatient, urban outpatient, and rural populations tested, respectively.

Conclusions Toxigenic *C. difficile* is an important but neglected aetiologic cause of infective diarrhoea in Central India. The higher prevalence within the urban inpatient setting likely reflects greater exposure to antibiotics and hospitalisation. Our findings underscore the need to enhance awareness of and testing of patients with diarrhoea in India, particularly in high-risk individuals with recent or ongoing antibiotic exposure or hospitalisation.

PTH-91 MULTIPLEX PCR FOR DETERMINING AETIOLOGY OF INFECTIOUS DIARRHOEA IN RURAL AND URBAN CENTRAL INDIAN POPULATIONS

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Introduction Infectious diarrhoea is a major cause of morbidity and mortality in Central India. There is an urgent unmet need to implement rapid point-of-care tests to deliver effective and targeted treatment plans. The aim of this exploratory study was to assess the performance of the FilmArray Gastrointestinal Panel for the detection of enteric pathogens directly from stool specimens collected from diarrhoeal and non-diarrhoeal control populations in Central India.

Methods Faecal samples were collected from participants with and without acute diarrhoea presenting to an inpatient or outpatient setting in Nagpur city district and rural Melghat. Each stool sample was stored at 4°C and preserved in Cary-Blair enteric transport medium for multiplex PCR using the FilmArray GI Panel according to the manufacturer's instructions. This panel allows for the simultaneous detection of 22 common diarrhoeal agents, including bacteria, viruses and protozoa. Baseline characteristics were also recorded and included: demographics, symptomatology, antibiotics exposure, duration of diarrhoea, hospitalisation status at recruitment, and duration, BMI, animal exposure, housing conditions, toilet access, and seasonality.

Results 179 participants provided stool samples for analysis on the FilmArray GI Panel. 70 and 109 participants were from rural Melghat and Nagpur urban district, respectively. Of these, 138 were from mainly non-hospitalised participants with acute diarrhoea from urban (n=89) and rural areas (n=49). In the urban cohort, 81% (88/109) of all diarrhoeal and non-diarrhoeal samples tested positive for one (27%) or more (54%) pathogens. In the rural cohort, a striking 97% (68/70) of samples yielded positivity to one (14%) or multiple organisms (83%). The most prevalent pathogen detected in both the diarrhoeal and control cohorts was *Enterohaemorrhagic E. coli* (51% vs 59%, respectively). However, other pathotypes of diarrhoeagenic *E. coli* were highly prevalent in both cohorts, including ETEC, EPEC, *Shigella*/EIEC, and STEC. A

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higher proportion of diarrhoeal samples tested positive to *Campylobacter* (12%) compared to the non-diarrhoeal control group (5%). Unlike the diarrhoeal samples, no control samples yielded positivity to *Vibrio cholerae*, *Cyclospora cayetanensis*, Astrovirus, Rotavirus A or Sapovirus

Conclusions Detection of high levels of polymicrobial enteric infections are prevalent in Central Indian symptomatic and asymptomatic populations. *E. coli* pathotypes predominate in both urban and rural settings. Further studies are required to understand the clinical significance of these mixed infections, as well as how best to manage them.

PTH-92 BRAIN-DERIVED NEUROTROPHIC FACTOR (BDNF) AND EPITHELIAL TO MESENCHYMAL TRANSITION (EMT), CLINICAL PERSPECTIVE IN COLORECTAL CANCERS

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Introduction Brain-Derived Neurotrophic Factor (BDNF), via its cognitive receptors Neurotrophic Receptor Tyrosine Kinases (NRTK/TrKs), is a neural survival factor and has been implicated in the development and progression of neurological cancer and selective non-neurological solid cancers. On cancer cells, BDNF has been reported to elicit biological influence on functions such as apoptosis, cellular motility and invasion, some of the hallmarks seen during Epithelial to Mesenchymal Transition (EMT). The present study examined the clinical and prognostic value of BDNF in connection with EMT markers, including Twist, Snail-1 (SNAI1), Slug (SNAI2), E(epithelial)-cadherin and N (neural)-cadherin, which were reported to have pivotal role in assessing disease progression and the clinical outcome of patients with certain solid tumours.

Methods A cohort of colorectal cancers were collected and analysed for the expression gene transcripts of Brain-Derived Neurotrophic Factor, together with a group of key EMTs biomarkers Twist, Snail-1, Slug, E-cadherin and N-cadherin. The clinical and prognostic values of the markers were collectively analysed in the connection with clinical and pathological factors and most importantly the clinical outcome of the patients.

Results The levels of BDNF transcript are significantly correlated with the Twist, Snail-1 and Slug ($p<0.001$, for all three molecules), to a lesser degree with E-cadherin ($p<0.01$) and marginally with N-cadherin. Whilst these markers had significant predictive effects on the clinical outcome of the patients, the combined analyses of BDNF and EMT markers together have offered a more significant power in predicting the overall survival of the patients. Patients with highly aberrant expression of BDNF/EMT markers showed a markedly shorter survival (40.7% vs 75% vs 100% survival rate over the 6 year follow-up period, respectively for those with high, moderate and low aberrant expression) (HR 2.7, $p<0.0001$). Similar correlation was seen between BDNF/EMT markers with the disease free survival (44.4%, 70% and 96.7% respectively, HR 2.07, $p=0.001$). These factors are in line with other clinical and pathological factors, including tumour staging ($p=0.027$) and Dukes staging ($p=0.017$), in the prediction of patient's survival.

Conclusions BDNF and the biochemical markers of EMT are aberrantly expressed in clinical colorectal cancer. The degree of aberration has a clear and significant clinical value in predicting the outcome of the patients.

Martin TA, Goyal A, Watkins G, *et al*. Ann. Surg. Oncol., 2005, 12: 488-496

PTH-93 AF6 (AFADIN/MLLT4) AND TIGHT JUNCTIONAL REGULATING SIPA1, EXPRESSION AND CLINICAL/ PROGNOSTIC VALUE IN COLORECTAL CANCER

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Introduction AF6 (Afadin, also known as MLLT4) is a junctional protein that belongs to the adhesion system of regulators of cell junctional adhesion. AF6 has been indicated in embryogenesis and development of certain types of haematological malignancies, and to a limited degree in solid tumours. In colorectal cancer, AF6 has been shown to regulate Cystic Fibrosis Transmembrane Conductance Regulator (CFTR). Recently, it was revealed that SIPA1 (Signal-Induced Proliferation-Associated-1), a kinase regulating cell cycle and mitogenesis, has recently discovered to regulate tight junctions. The present study explored the expression and connection between AF6 and SIPA1 in human colorectal cancers.

Methods The expression levels of AF6 and SIPA1 in human colorectal tumour and normal tissues were quantitatively determined by way of gene transcript analyses. The expression was analysed against the clinical and pathological parameters, together with clinical outcome of the patients including recurrence, metastasis and colorectal related death. Statistical methods were Mann Whitney U test for comparisons, logistic regression and Kaplan-Meier's methods for survival analyses.

Results Both AF6 and SIPA1 had an aberrant pattern of expression in colorectal tumours compared with normal colon control tissues. Our dataset surprisingly displayed a highly significant correlation between AF6 and SIPA1 ($r=0.904$, $p<0.00001$). AF6 and SIPA1 had significant value in predicting overall survival (Hazard Ratio (HR) 3.7, $p=0.05$, for AF6 and HR 4.8, $p=0.01$) and relapse free survival ($p=0.044$ and $p=0.016$ for AF6 and SIPA1 respectively). When the expression pattern of AF6 and SIPA1 were integrated for analyses, they further enhanced the prediction power for both overall survival (mean survival 38.5 months, 111.6 months and 159.6 months respectively for the differential expression pattern, $p=0.002$) and relapse free survival (mean survival 32.3, 108.4 and 153.3 months respectively, $p=0.005$). AF6/SIPA1 expression pattern is an independent prognostic factor for overall survival ($p=0.008$), in contrast to staging, differentiation and nodal status which showed either independency in the current cohort. AF6/SIPA1 is a weak factor for disease free survival ($p=0.06$), in contrast to tumour staging and nodal status which had independent predictive value ($p<0.05$).

Conclusion AF6, together with SIPA1, are aberrantly expression in clinical colorectal cancer and have significant values in predicting the progression and the outcome of the patients. The molecules are potential targets in colorectal cancer.

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