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Ulcerative colitis rodent model treatment with opium tincture protocol, a clinical and gene expression profiling study

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Abstract

Ulcerative Colitis (UC) is a chronic, idiopathic inflammatory bowel disease characteristically restricted to the colonic mucosa. While the precise etiology of UC remains to be fully elucidated, a complex interplay of genetic predisposition and autoimmune responses drives persistent mucosal inflammation and structural injury. The present study report the effects of a new protocol of oral consumption of opium tincture taper up off on gene expression profile of UC models of rats using microarray.

The rat model of UC produced using Dextran Sulfate Sodium (DSS)-Induced acute colitis. The UC model rats treated with a novel taper up off opium tincture oral administration protocol called Dezhakam-Step-Time (DST) in four different dosages. Clinical situation of UC models have examined with disease activity index. The colon tissues collected and RNA extraction and cDNA synthesis performed. The expression profiling evaluated using microarray followed by the quantitative Real-time PCR.

Gene expression profiling revealed that 1138 genes found differentially expressed in UC models vs. normal controls. The UC model Differentially Expressed Genes (DEGs) are clustered in immune system, inflammation mechanisms, structural and cellular remodeling, modulation of ulcer healing and metabolic pathways associated to cancer. The taper up off opium tincture oral administration showed strong down regulation of inflammation and antigen processing and presentation pathways including TNF, JAK-STAT and cytokine signaling related genes along with up regulation of wound healing-related genes. Best results observed in intermediate dosages.

The findings of the gene expression profiling study regarding UC model rats treated with taper up off opium tincture suggest that the new protocol may induce the reduction of the inflammation along with the activation of wound healing which may lead to decrease of severity of disease and potential repair of injured tissues such as intestinal mucus respectively. While the addictive effect of opium is a major health concern, the DST protocol originally had been designed for the opium addition treatment. Given that, it seems that the addiction free administration of opium tincture could be a new hope for reduction of symptoms severity and the risk of UC induced colon cancer in UC patients.

Introduction

Ulcerative Colitis (UC) is a complex inflammatory bowel disease characterized by chronic inflammation of the colon's mucosa with unclarified etiology. The age of onset of UC, mostly occurring in two periods of age, between 15-25 years and between 55-65 years of age [1]. The UC prevalence has higher rates in North America and Europe compared to Asia and Africa, although UC prevalence is rising worldwide [2]. In Ulcerative Colitis (UC), the integrity of the intestinal mucosal barrier compromised, frequently coinciding with significant gut microbiota dysbiosis. This barrier disruption facilitates a robust inflammatory cascade characterized by the extensive infiltration of immune cells into the mucosa and submucosa. Consequently, achieving robust mucosal healing is a primary therapeutic objective essential for tissue repair and the restoration of physiological homeostasis [3]. Clinically, UC typically presents as a constellation of symptoms, including hematochezia, increased bowel frequency, abdominal tenderness, and systemic manifestations such as weight loss, pyrexia, and fatigue [4].

The pathophysiology of UC is multifactorial and unclarified but the genetic risk factors along with environmental factors and immune dysregulation have been consider as involving factors [5]. Multiple Genome Wide association studies indicate that the main genetics risk factor including candidate genes and epigenetic modifications are associated with inflammation, immune regulation and epithelial permeability [6]. Mucosal regeneration in Ulcerative Colitis (UC) governed by a complex interplay of molecular signaling and microbial ecology. Specifically, the activation of STAT3, often mediated via IL-22 signaling, represents a critical pathway for effective epithelial repair. This process further augmented by CARD9, which appears to facilitate intestinal healing through the downstream regulation of IL-22 [7]. Conversely, the maintenance of microbial homeostasis is paramount; a shift toward dysbiosis—characterized by the depletion of commensal species and the proliferation of pathobionts—can significantly impede the restorative process [8]. The machine learning analysis on several gene expression profiling studies conducted on UC patients showed most of the differentially expressed genes belong to chronic inflammation of the colon, predominantly driven by an aberrant immune response activated dendritic immune cells, antigen presentation and initiation of adaptive immune responses. In addition, innate immunity, and bacteriostatic mechanisms relate gene had affected in UC patients [9].

Beyond genetics, environmental influences—specifically changes in the gut microbiome—and lifestyle factors such as smoking and diet play a role in the disease. In genetically predisposed cohorts, an atypical immune reaction to luminal antigens precipitates persistent inflammation and subsequent mucosal injury [10].

Although opium-derived therapeutics are established analgesics, their clinical application necessitates rigorous oversight due to the risk of adverse effects and physiological dependence. Opium tincture (laudanum) is a multifaceted opioid analgesic traditionally utilized for the management of severe diarrhea, neonatal opioid withdrawal syndrome, and antitussive therapy [11]. Despite its long-standing use, the impact of opium and its constituent alkaloids on mammalian transcriptomic profiles and epigenetic landscapes remains poorly defined. Highthroughput gene expression analysis may provide critical insights into the pleiotropic effects of opioid compounds on systemic physiological processes, including the modulation of immune responses and the molecular mechanisms governing wound repair [12].

Previously we reported the early report that focuses on the expression level of wound healingrelated genes in opium tincture-treated, untreated, and control groups. The present article is the report of the clinical and molecular study of the ulcerative colitis rat model treated with a novel protocol of taper up off opium tincture to evaluate the transcriptomic pattern of opium on ulcerative colitis.

Materials and methods

Animal modeling protocol

This study utilized Sprague-Dawley rats, with twelve males and twelve females aged 7–8 weeks in each group. The body weights of male ranged from 220 grams to 245 grams, and female body weights ranged from 192 grams to 202 grams in the first day of experiment. Due to similarity to the pathology of human UC, we used the Dextran Sulfate Sodium (DSS) Induced acute Colitis [13]. The 3% DSS administered in drinking water for seven days to induce the acute colitis. Mice given normal drinking water for 5 days before starting the treatment after 12 days. Animals housed in groups of three per cage with access to ad libitum food. The daily body weights and morbidity had measured each morning and fecal samples collected from all rats on sacrifice day. Disease Activity Index (DAI) scores evaluated based on body weight recovery at sacrifice, stool consistency, and the presence of blood in stool. Histopathological damage scored from H&E-stained distal colon cross-sections, evaluating crypt architecture, inflammatory cell infiltrates, muscle thickening, goblet cell depletion, and crypt abscesses, as previously described [14].

Treatment protocol with opium tincture

Treatment with opium tincture administered to rats, according to a laboratory-designed taperup-off protocol, the Dezhakam-Step-Time (DST) method. The method consisted of an 18-step administration schedule, and doses given twice daily for 9 days to taper up until reaching to highest dosage, and then twice daily for another 9 days to taper down until taper off the administration. On the other words, treatment start with the lowest dose, which then increased by 20% (multiplied by a coefficient of 0.8) at each administration. After 18 administrations during nine days, the process reversed, with the dose reduction by 20% at each of the following 18 administrations. Consequently, in this protocol, the first and last doses would be the same and lead to termination of usage. The outline of experimental groups and group's description presented in Table 1. Four different dosage regimens have programmed, the first regimen started at 2.35 mg/kg, reaching 65.58 mg/kg by the 18th dose. The starting doses for the second, third, and fourth regimens were 3.52 mg/kg, 4.7 mg/kg, and 5.87 mg/kg, respectively.

Scarification and tissue collection

At the end of the experiment, rodents were euthanized using overdose of sodium pentobarbital and administered intravenously based on protocols published in previous pharmacology and toxicity studies. Then about 2-centimeter portion of the distal colon of euthanized rats was collected for molecular analysis. Both housing and euthanasia procedures performed based on the ARRIVE55 Guidelines checklist and protocols of previous studies [15,16].

RNA extraction and cDNA synthesis

The RNA extraction process from the colon tissues performed using the mini kit (Qiagen, Hilden, Germany) according to the manufacturer's instructions. Due to the high rate of DNA contamination of tissues extracted RNAs, the extracted RNA was treated with DNase I, RNasefree (Fermentas, Latvia; #EN0521) following the manufacturer's protocol. The quantity and quality of the extracted RNA assessed using the Nanodrop-1000 spectrophotometer, and the BioRad Experion automated electrophoresis system (BioRad Laboratories Inc.) respectively. The main extracted RNA samples were stored at under -80° centigrade degree freezer until further processing. The cDNA was synthesized using the RevertAid Premium First Strand cDNA Synthesis Kit (Thermo Scientific - Fermentas, Latvia; #K1652), following the manufacturers protocol.

Transcriptomic profiling via DNA microarray

Whole-genome expression analysis was performed using the Affymetrix GeneChip™ Rat Genome 230 2.0 Array, a platform optimized for enabling us to conduct an unbiased assessment in rat models. All procedures, including aRNA target labeling, fragmentation, hybridization, and scanning, adhered to the manufacturer's standardized protocols (Affymetrix, Santa Clara, CA).

Initial sample integrity verified via Agilent Bioanalyzer Quality Control (QC). Subsequently, 100 ng of total RNA per sample processed using the GeneChip 3' IVT Express Kit, involving reverse transcription to double-stranded cDNA followed by biotin-labeled transcription. The resulting fragmented aRNA hybridized to the arrays for 16 hours at 45°C. Post-hybridization, arrays washed out and stained using the GeneChip Hybridization, Wash, and Stain Kit on a GeneChip Fluidics Station 450. Finally, the chips scanned using the Affymetrix GeneChip Scanner 3000, with all arrays successfully meeting established QC metrics.

Bioinformatics and statistical data processing

Microarray data were processed using Microarray Analysis Suite (MAS) 5.0, Data Mining Tool 2.0, and Genesis 2.0 (GeneLogic Inc., Gaithersburg, MD, USA). Signal intensities for all represented genes normalized and scaled to a target intensity of 100. To mitigate false-positive results, MAS 5.0 employed for initial filtering, and the remaining dataset analyzed via Genesis 2.0. Differentially expressed genes (DEGs) defined as those exhibiting a Fold-Change (FC) >2.0 with a statistically significant adjusted P-value, determined after applying the Bonferroni correction for multiple comparisons.

Functional enrichment and pathway analysis

Functional annotation & Gene Ontology (GO) assessments performed for the final subset of DEGs that demonstrated consistent expression patterns across both microarray and Real-time PCR validations. Integrated enrichment algorithms from the Database for Annotation, Visualization, and Integrated Discovery (DAVID, v6.8) and Strand Genomics (Redwood City, CA, USA) utilized for functional clustering. The biological pathway mapping conducted using the Kyoto Encyclopedia of Genes and Genomes (KEGG) enrichment tool. Comparative visualizations of DEGs, including GO terms and KEGG pathways, were generated as Venn diagrams using the Bioinformatics and Evolutionary Genomics suite (Van de Peer Lab).

DEGs results confirmation with quantitative Real-Time PCR (qPCR)

To validate the transcriptomic data, all significantly differentially expressed genes identified via microarray subjected to mRNA level evaluation with Quantitative Real-time PCR (qPCR). Standard curves were generated using serial dilutions (1:4) of pooled cDNA derived from randomly selected control rat samples. Gene-specific primers and probes were designed utilizing MEGA7 software and cross-referenced for specificity using the NCBI BLAST-Primer tool. Quality checking criteria of qPCR assessments included an R2 value that should be more than 0.99 in the standard curve and no signal detection for No-Template Control samples (NTC). Amplification efficiency was verified using Lin-Reg PCR software (Amsterdam, Netherlands). Reactions executed using the TaqMan® PCR Starter Kit (Thermo Scientific - Fermentas, Latvia) on a CFX96 Touch Real-Time PCR Detection System (Bio-Rad, CA, USA). Relative expression ratios calculated according to the Pfaffl formula.

Statistical analysis

Statistical processing performed using SPSS version 25.0. Data normality examined via the Kolmogorov-Smirnov test. Comparative analysis between multiple experimental groups conducted using one-way ANOVA. To control for potential confounding factors, ANCOVA used to evaluate the persistence of significant differences between groups, with RNA integrity number, cDNA synthesis quality, qPCR plates/runs, and primer and probe efficiency defined as covariates. To mitigate the risk of Type I errors, the Bonferroni correction test was applied for all post-hoc multiple comparisons. Descriptive statistics have reported as mean ± standard deviation (SD).

Results

Clinical observations

To assess the clinical situations of animal models, the Disease Activity Index (DAI) utilized, incorporating parameters of weight loss, stool consistency, and the presence of gross fecal blood. Significant reduction in body weight (P=0.003), along with clinical manifestations of acute colitis were observed in all DSS-treated animals. DSS-rats showed hematochezia and diarrheic stools. Furthermore, subjects exhibited behavioral signs of abdominal discomfort and systemic malaise, including piloerection and reduced locomotor activity. These clinical markers peaked during the acute phase of induction and showed varying degrees of resolution. These observations align with the characteristic phenotypic profile of the DSS model, confirming the successful induction of mucosal injury prior to therapeutic intervention. The locomotor activity improved in G3 and G4 groups during and after the treatment. During the treatment period, one female rat from G3, and two male and one female rat from the untreated DSS-induced colitis group died before the scarification day.

Gene expression profiling findings

The RNA quality evaluation determined all samples had RNA Quality Indicator (RQI) values higher than 9.5. Numerus amount of genes differentially expressed in all DSS-induced colitis models compared with Normal and Sham groups. A summary of these statistically significant DEGs have provided in Table 2, with their overlapping and distinct expression patterns visualized via Venn diagrams in Figure 1. Pathway enrichment analysis revealed significant pathways alteration across all DSS-induced colitis models, regardless of treatment status. While a core set of shared DEGs have identified across all opium-treated cohorts, distinct gene expression signatures were unique to specific tapering protocols. Notably, the DST3 and DST4 protocols elicited significant modulation of genes associated with tumor suppression and protooncogene regulation, as well as transcription factors governing interleukin production and metabolic homeostasis. Furthermore, treatment groups demonstrated robust alterations in pathways related to innate immunity, Natural Killer (NK) cell regulation, JAK-STAT, TNF signaling cascades, regulation of intracellular signal transduction, wound healing and inhibition of metastasis. The statistical outcomes of the pathway enrichment analysis, comparing the various treatment cohorts against the control groups, summarized in Table 3 and illustrated via Venn diagrams in the Figure 2. Notably, no significantly DEGs identified between the normal and sham groups, confirming that the experimental procedures themselves did not introduce confounding transcriptomic variations.

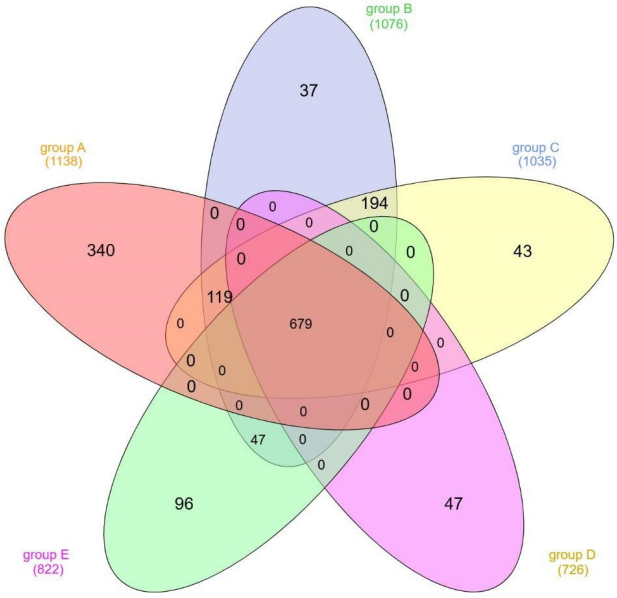

Figure 1: Venn diagram of differentially expressed genes based on microarray analysis of UC model and treatment groups. **(Group A)** DSS-induced colitis model with no treatment (red) **(Group B)** DSS-induced colitis model treated with protocol 1 opium tincture (blue), **(Group C)** DSS-induced colitis model treated with protocol 2 opium tincture (yellow), **(Group D)** DSS induced colitis model treated with protocol 3 opium tincture (purple), **(Group E)** DSS-induced colitis model treated with protocol 4 opium tincture (green).

The most significantly altered pathways in group A were including immune system signaling

(JAK-STAT Signaling Pathway, TNF Signaling Pathway, and Cytokine-Cytokine Receptor Interaction), Epithelial/ Mesenchymal Transition and Extracellular Matrix Remodeling, TollLike Receptor Signaling, Antigen Processing and Presentation, bioenergetics and mitochondria signaling, matrix metalloproteinase regulation and cell matrix adhesion. In treatment groups the most significantly down regulated genes in groups D and E were found in TNF signaling pathway (p=0.00003 in group D and p=0.00004 in-group E), JAK-STAT signaling pathway (p=0.00003 in-group D, p=0.00003 in-group E) and Cytokine-Cytokine Receptor Interaction signaling pathway (p=0.00003 in group D and p=0.00004 in-group E). In addition, the most significantly up regulated genes in groups D and E were found in bioenergetics and mitochondria signaling pathway (p=0.00003 in-group D, p=0.00003 in-group E).

No significant difference observed in mRNA levels of any genes between female and male rats in any group. To corroborate the transcriptomic profiles obtained via microarray, the mRNA expression levels of all identified Differentially Expressed Genes (DEGs) validated using qPCR. The qPCR findings were highly concordant with the microarray data across all groups, exhibiting consistent trends in both the directionality and the magnitude of expression changes.

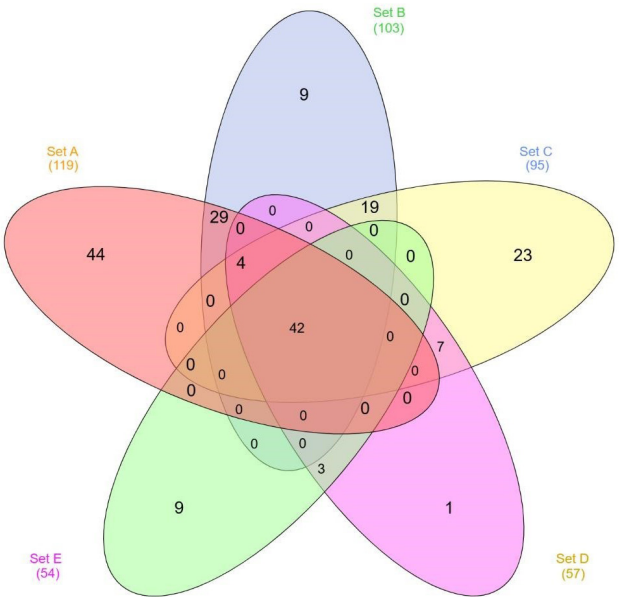

Figure 2: Venn diagram of enriched pathway analysis based on differentially expressed genes in UC model and treatment groups. **(Group A)** DSS-induced colitis model with no treatment (red), **(Group B)** DSS-induced colitis model treated with protocol 1 opium tincture (blue), **(Group C)** DSS-induced colitis model treated with protocol 2 opium tincture (yellow), **(Group D)** DSSinduced colitis model treated with protocol 3 opium tincture (purple), **(Group E)** DSS-induced colitis model treated with protocol 4 opium tincture (green).

Table 1: Description of experimental groups.

Group name	Description of group
A	DSS-induced colitis model with no treatment
B	DSS-induced colitis model treated with protocol 1 opium tincture
C	DSS-induced colitis model treated with protocol 2 opium tincture
D	DSS-induced colitis model treated with protocol 3 opium tincture
E	DSS-induced colitis model treated with protocol 4 opium tincture
F	DSS-induced colitis model with force-feeding of water (sham group)
G	normal rats with no treatment (control group)

All treatment protocols have based on Dezhakam-step-time method with different dosage of opium tincture. Protocol 1: 2.35 mg/kg force-feeding of opium tincture as first dosage, Protocol 2: 3.52 mg/kg force-feeding of opium tincture as first dosage, Protocol 3: 4.7 mg/kg forcefeeding of opium tincture as first dosage, Protocol 4: 5.87 mg/kg force-feeding of opiumtincture as first dosage. Each group was including twelve male and twelve female rats

Table 2: Differentially expressed genes (Fold-Change >2.0 and P<0.05) results of microarray assessments in comparison of each group with group G.

Groups compared with G group	Total DEGs	Up-regulated genes	Down-regulated genes
A	1138	674	464
B	1076	633	443
C	1035	452	583
D	726	277	449
E	822	310	512
F	1144	688	456

Table 3: Enriched pathway analysis in each group.

Number	Group comparisons	Number of significantly altered pathways
1	A vs. G	119
2	B vs. G	103
3	C vs. G	95
4	D vs. G	57
5	E vs. G	54
6	F vs. G	110

Discussion

Transcriptomic analysis revealed that 1138 genes found differentially expressed in the DSS induced colitis model compared to healthy controls. These DEGs have clustered within pathways related to the inflammatory response, cell adhesion system, and bioenergetics mechanisms, underscoring the systemic molecular disruption caused by mucosal injury.

It seems that DST protocol effects on DEGs could be dose dependent at some pathways such as immune system, wound healing or bioenergetics pathways and not dose dependent at some other pathways associated to immune system. For example, two shared DEGs in all four treatment groups with similar expression level, were genes of OLFM4 and C4BPB which were found related to six immune cells, namely Macrophages M1, Macrophages M2, Mast cells activated, Mast cells resting, Monocytes, and Natural killer cells activated [9].

Results of shared DEGs in treatment groups showed that several genes in wound healing related pathways such as epithelial to mesenchymal transition and extracellular matrix remodeling were down-expressed in UC models and upregulated during the treatment of all four groups while their activation was mostly dose-dependent. On the other hand, numerous genes related to the immune system including IL22, IL1b, and SERPINE1 were highly overexpressed in UC models and were downregulated in treatment group that may indicate that DST protocol reduce the UC-induced inflammation [17] but the down expression level were completely dose dependent. These dosage dependent differences mostly detected in JAKSTAT signaling pathway, TNF Signaling Pathway and Cytokine-Cytokine Receptor Interaction reflects. JAK-STAT signaling pathway is central to the inflammatory cascade and cells response to cytokines. The TNF Signaling Pathway is involved in primary driver of mucosal damage and a key target for clinical treatments and the Cytokine-Cytokine Receptor Interaction reflects, is the massive cytokine storm and leukocyte infiltration into the mucosa and submucosa [18].

Mucosal wound healing in UC models involves a complex interplay between immune inflammatory regulators and structural remodeling factors. The IL-22/STAT3 axis, modulated by SERPINE1 and CARD9 related pathways, serves as the primary engine for epithelial proliferation and barrier restoration [19]. This framework is structurally and angiogenically supported by downstream cascades—including iNOS/VEGF-mediated revascularization, cell adhesion, and FGF/MAPK-induced cellular proliferation. The regulatory role from TIMP2, CXCL1, PDGFRA, and BMP2, combined with R-SMAD activity pathways form a highly integrated network that drives tissue regeneration and resolves colitis-induced mucosal injury [20-28]. All of the mentioned pathways have detected and found activated in pathway analysis of DEGs in all DST treated groups.

Based on previous studies, there is an association between gene expression signature and disease severity of UC patients [29]. Two cluster of pathways indicated in UC severity expression signature are antigen processing presentation, and toll-like receptor signaling [30]. Enriched analysis revealed up regulation of DEGs in these pathways in group A as well as down expression of several DEGs in groups D and E. The expression level of genes involved in antigen processing and presentation in-group D were dramatically decreased, near to group G, which may indicate the less pathological adaptive immune response by strong reduction of self-sustaining loop of priming adaptive T-cell responses in the colonic tissue. In addition, the toll-like receptor signaling down regulation, determined in-group D may lead to mitigate the inflammation, activation of damaged microbiota, and improve the gut function. Prior investigations have indicated that the Dezhakam-Step-Time (DST) method a standardized tapering protocol may mitigate the risk of opioid dependence, offering a potentially viable strategy for utilizing the therapeutic properties of opium tincture [31,32]. The chronic epithelial damage and accelerated cell turnover inherent to sustained UC represent wellestablished risk factors for the development of colitis-associated colorectal malignancy. Therapeutic strategies shall focus dualistically on the robust attenuation of the inflammatory cascade and the concurrent promotion of epithelial wound healing as well to restore mucosal barrier integrity and achieve long-term clinical remission. The specific transcriptomic and physiological impacts of the DST protocol on Ulcerative Colitis (UC) especially due to inflammatory cascades and tissue regeneration is not

fully elucidated, and the potential addictive effect of opium tincture remain a concern regardless of DST taper off strategy. Nevertheless, the present study findings may suggest that this novel oral administration regimen may attenuate colonic inflammation. Furthermore, our data indicate a concomitant activation of mucosal wound healing processes, which may collectively promote disease remission and potentially reduce the long-term risk of colitis-associated colorectal malignancy.

References

1. O'Shea MD, Kaplan GG. Epidemiology of inflammatory bowel disease. Gastroenterol Clin North Am. 2016; 45: 147-158.
2. Molodecky NA, Soon IS, Rabi DM, Ghali WA, Ferris M, Chernoff G, et al. Increasing incidence and prevalence of the inflammatory bowel diseases with time, based on systematic review. Gastroenterology. 2012; 142: 46-54.e42.
3. Pavlidis P, Tsakmaki A, Pantazi E, Li K, Cozzetto D, Digby-Bell J, et al. Interleukin-22 regulates neutrophil recruitment in ulcerative colitis and is associated with resistance to ustekinumab therapy. Nat Commun. 2022; 13: 5820.
4. Harbord M, Eliakim R, Bettenworth D, Karmiris K, Katsanos K, Kopylov U, et al. Third European evidence-based consensus on diagnosis and management of ulcerative colitis. Part 2: current management. J Crohns Colitis. 2017; 11: 769-784.
5. O'Shea MD, Kaplan GG. Epidemiology of inflammatory bowel disease. Gastroenterol Clin North Am. 2016; 45: 147-158.
6. Molodecky NA, Soon IS, Rabi DM, Ghali WA, Ferris M, Chernoff G, et al. Increasing incidence and prevalence of the inflammatory bowel diseases with time, based on systematic review. Gastroenterology. 2012; 142: 46-54.e42.
7. Bai J, Bai J, Yang M. Interleukin-22 attenuates acute pancreatitis-associated intestinal mucosa injury in mice via STAT3 activation. Gut Liver. 2021; 15: 771.
8. Çavdar M, Çavdar M. Alternative therapeutic applications used in the treatment of ulcerative colitis: probiotics, prebiotics, synbiotics and fecal microbiota transplantation. Prog Nutr. 2023; 25.
9. Wang J, Li L, Chen P, He C, Niu X. Exploration and verification a 13-gene diagnostic framework for ulcerative colitis across multiple platforms via machine learning algorithms. Sci Rep. 2024; 14: 15009.
10. Ungaro R, Bernstein CN, Gearry RB, Hobert AM, Ng SC, Annese V, et al. Ulcerative colitis: pathophysiology, diagnosis, and management. Nat Rev Gastroenterol Hepatol. 2017; 14: 690-707.
11. Danne C, Lamas B, Lavelle A, Michel ML, Da Costa G, Pham HP, et al. Dissecting the respective roles of microbiota and host genetics in the susceptibility of Card9−/− mice to colitis. Microbiome. 2024; 12: 76.
12. Liu T, Guo Y, Liao Y, Liu J. Mechanism-guided fine-tuned microbiome potentiates antitumor immunity in HCC. Front Immunol. 2023; 14: 1333864.
13. Okdahl T, Høyer KL, Knoph CS, Davidsen L, Larsen IM, Mark EB, et al. Opium tincture has anti-propulsive effects in patients with chronic diarrhea: a randomized, placebo-controlled, and cross-over trial. Scand J Gastroenterol. 2024; 59: 1023-1034.
14. Dezhakam H, Dezhakam A, Dezhakam A, Dezhakam S, Haghighatfard A. Short report of potential myelinogenesis effects of taper up-off of opium tincture in rodent model of multiple sclerosis. J Neurol Neurol Sci Disord. 2024; 10: 21-26.
15. Kiesler P, Fuss IJ, Strober W. Experimental models of inflammatory bowel diseases. Cell Mol Gastroenterol Hepatol. 2015; 1: 154-170.
16. Tusé D, Reeves M, Royal J, Hamorsky KT, Ng H, Arolfo M, et al. Pharmacokinetics and safety studies in rodent models support development of EPICERTIN as a novel topical wound-healing biologic for ulcerative colitis. J Pharmacol Exp Ther. 2022; 380: 162-170.
17. Yang T, Ma X, Wang R, Liu H, Wei S, Jing M, et al. Berberine inhibits IFN-γ signaling pathway in DSS-induced ulcerative colitis. Saudi Pharm J. 2022; 30: 764-778.
18. Zhang L, Mao R, Lau CT, Chung WC, Chan JC, Liang F, et al. Identification of useful genes from multiple microarrays for ulcerative colitis diagnosis based on machine learning methods. Sci Rep. 2022; 12: 9962.
19. Shomer NH, Allen-Worthington KH, Hickman DL, Jonnalagadda M, Newsome JT, Slate AR, et al. Review of rodent euthanasia methods. J Am Assoc Lab Anim Sci. 2020; 59: 242-253.
20. Zhu F, Yang T, Ning M, Liu Y, Xia W, Fu Y, et al. MiR146a alleviates inflammatory bowel disease in mice through systematic regulation of multiple genetic networks. Front Immunol. 2024; 15: 1366319.
21. Martín Adrados B. Intestinal epithelial cell response to endoplasmic reticulum stress in inflammatory bowel disease.
22. Jian F, Yanhong J, Limeng W, Guoping N, Yiqing T, Hao L, et al. TIMP2 is associated with prognosis and immune infiltrates of gastric and colon cancer. Int Immunopharmacol. 2022; 110: 109008.
23. Sakthivel KM, Guruvayoorappan C. Amentoflavone inhibits iNOS, COX-2 expression and modulates cytokine profile, NF-κB signal transduction pathways in rats with ulcerative colitis. Int Immunopharmacol. 2013; 17: 907-916.
24. Ou W, Xu W, Liu F, Guo Y, Huang Z, Feng T, et al. Increased expression of yes-associated protein/YAP and transcriptional coactivator with PDZ-binding motif/TAZ activates intestinal fibroblasts to promote intestinal obstruction in Crohn's disease. EBioMedicine. 2021; 69.
25. Liu J, Xiao Q, Xiao J, Niu C, Li Y, Zhang X, et al. Wnt/β-catenin signalling: function, biological mechanisms, and therapeutic opportunities. Signal Transduct Target Ther. 2022; 7: 3.