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# 思连康与美沙拉秦对溃疡性结肠炎患者肠黏膜 TLR4、NF-κb 表达与肠道菌群的影响 *

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**摘要 目的：**探讨思连康联合美沙拉秦用于溃疡性结肠炎的疗效及对肠道黏膜肠黏膜 Toll 样受体 (TLR)4、核转录因子(NF)-κb 表达与肠道菌群的影响。**方法：**选择 2015 年 1 月至 2017 年 1 月我院接诊的 92 例溃疡性结肠炎患者，通过随机数表法分为观察组(n=46)和对照组(n=46)，对照组给予美沙拉秦肠溶片治疗，观察组联合思连康治疗，均连续治疗 6 周。比较两组治疗前后临床症状评分、Baron 内镜评分、肠粘膜 TLR4、NF-κb 表达、肠道菌群数量的变化及不良反应的发生情况。**结果：**治疗后，两组临床症状评分、Baron 内镜评分均较治疗前均明显降低 (P＜0.05)，且观察组腹痛、腹泻、脓血便及 Baron 内镜评分均明显低于对照组 (P＜0.05)；两组肠粘膜 TLR4、NF-κb 的表达较治疗前均明显降低(P＜0.05)，且观察组肠粘膜 TLR4、NF-κb 表达均明显低于对照组(P＜0.05)；两组双歧杆菌、乳杆菌、酵母菌、梭菌数量较治疗前均显著改变(P＜0.05)，且观察组双歧杆菌、乳杆菌数量明显多于对照组，酵母菌、梭菌数量明显少于对照组(P＜0.05)；两组治疗过程中均未有明显不良反应，差异无统计学意义(P＞0.05)。**结论：**思连康联合美沙拉秦治疗溃疡性结肠炎的临床效果显著，可有效缓解临床症状，促进粘膜修复，其内在机制可能和降低肠粘膜 TLR4、NF-κb 的表达及调节肠道微生态平衡相关。

**关键词：**溃疡性结肠炎；思连康；美沙拉秦；Toll 样受体 4；核转录因子 -κb

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# Clinical Efficacy of Siliankang Combining with Mesalazine in Treatment of Ulcerative Colitis and Effects on Intestinal Mucosa TLR4 and NF-κb Expressions and Intestinal Microflora*

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**ABSTRACT Objective:** To study the curative efficacy of siliankang combining with mesalazine in the treatment of ulcerative colitis and effects on the intestinal mucosa toll like receptor (TLR4), nuclear factor (NF-κb) expressions and intestinal microflora. **Methods:** 92 patients with ulcerative colitis who were treated from January 2015 to January 2017 in our hospital were selected and divided into the observation group (n=46) and the control group (n=46) according to random number table. The control group was treated with mesalazine, while the observation group was combined with siliankang, they were continuously treated for 6 weeks. The changes of clinical symptom score, Baron endoscopy score, intestinal mucosal TLR4 and NF-κb expressions and intestinal flora before and after treatment as well as the incidence of adverse events were compared between the two groups. **Results:** After treatment, the clinical symptom score and Baron endoscopic score of both groups were significantly lower than those before treatment (P＜0.05); the bellyache, diarrhea, pus and blood stool and endoscopic Baron score in the observation group were significantly lower than those of the control group (P＜0.05); the intestinal mucosal TLR4 and NF-κb expressions of both groups were significantly lower than those before treatment (P＜0.05); the intestinal mucosal TLR4 and NF-κb expressions in the observation group were significantly lower than those of the control group (P＜0.05); the number of *bacillus bifidus*, *lactobacilli*, *microzyme* and *clostridium* of both groups was significantly changed compared with those before treatment (P＜0.05); the number of *bacillus bifidus*, and *lactobacilli* of observation group were significantly higher than those of the control group, the number of microzyme and clostridium were significantly lower than those of the control group(P＜0.05); there was no obvious adverse reaction in the two groups (P＞0.05). **Conclusion:** Siliankang combined with mesalazine can effectively relieve the clinical symptoms and promote mucosal repair of patients with ulcerative colitis, the underlying mechanism may be related to the reduction of intestinal mucosal TLR4 and NF-κb expressions and the regulation the intestinal microecological balance.

**Key words:** Ulcerative colitis; Siliankang; Mesalazine; Toll like receptor 4; Nuclear factor-κb

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## 前言

溃疡性结肠炎是一种结肠粘膜层所发生的非特异性炎性病变,表现为溃疡糜烂,多累及远端结肠,严重的甚至累及全结肠,患者以腹痛、腹泻、脓血便等为主要临床症状,近年来该病的发生率呈上升趋势[1,2]。研究表明机体免疫异常在该病中起着重要作用,Toll 样受体(TLR)是种跨膜信号转导受体,可通过识别并结合病原识别模式分子激活核转录因子(NF)-κb,导致炎症因子产生增加, 进而介导肠粘膜内的炎症反应和免疫反应。此外,肠道菌群失调和该病的发生、发展也存在着密切的关系[3,4]。目前,该病的治疗以氨基水杨酸类药物如美沙拉秦等为主,对缓解肠道炎症具有一定作用[5]。近年来,国内外均有学者提出补充益生菌有助于改善溃疡性结肠炎患者的肠道菌群失衡,继而达到缓解病情的效果[6,7]。本研究主要探讨了思连康联合美沙拉秦对溃疡性结肠炎肠粘膜 TLR4、NF-κb 的影响,旨在为思连康联合美沙拉秦的临床应用提供更多的理论支持,现将结果报道如下。

## 1 资料与方法

### 1.1 一般资料

选择 2015 年 1 月至 2017 年 1 月我院接诊的 92 例溃疡性结肠炎患者。纳入标准[8]:① 符合溃疡性结肠炎诊断标准,并通过内镜检查得以确诊;② 知情同意此次研究。排除标准[9]:① 合并感染性肠炎、放射性及缺血性肠炎等;② 近期服用过免疫抑制剂、皮质激素等;③ 合并全身免疫性疾病;④ 对研究药物过敏。以随机数表法分为两组,每组 46 例。观察组男 25 例,女 21 例;年龄 30~56 岁,平均(43.25± 3.16)岁;轻度 32 例,中度 14 例。对照组男 23 例,女 23 例;年龄 32~57 岁,平均(43.40± 3.04)岁;轻度 30 例,中度 16 例。两组一般资料比较差异均无统计学意义(P>0.05),具有可比性。

### 1.2 治疗方法

对照组给予美沙拉秦肠溶片(规格 0.5 g,厂家:德国 Losan Pharma GmbH,国药准字 H20150124),0.5 g/ 次,3 次 /d。观察组联合思连康(规格 0.5 g,厂家:杭州远大生物制药有限公司,国药准字 S20060010),0.5 g/ 次,3 次 /d。连续治疗 6 周,两组治疗过程中均忌食辛辣、海鲜等刺激类易加重病情的食物。

### 1.3 观察指标

**1.3.1 临床症状评分** 无腹痛、无腹泻、无脓血便记 0 分;伴有轻微腹痛、每日腹泻<4 次、少量脓血便记 3 分;腹痛频繁、每日腹泻次数 4~6 次、脓血便较多记录 6 分。

**1.3.2 Baron 内镜评分** 粘膜正常为记 0 分;粘膜充血、血管纹理模糊记录 1 分;粘膜存在接触性出血记录 2 分;粘膜存在自发性出血记录 3 分;粘膜有大小不等溃疡且合并出血记录 4 分。

**1.3.3 肠粘膜 TLR4、NF-κb 的表达** 治疗前后,使用结肠镜钳取肠粘膜组织,常规石蜡包埋、切片及 HE 染色,使用常规免疫组化 SP 发对 TLR4、NF-κb 的表达进行检测, 其中 TLR4 多克隆抗体、NF-κb 单克隆抗体均购于美国 Santa Cruz 公司, 所有标本均使用 Motic Med6.0 数码医学图像分析系统, 若细胞质染为棕黄色则为阳性细胞,对阳性细胞染色的积分光密度进行计算,结果以 OD 值表示。

**1.3.4 肠道菌群检测** 治疗前后, 采取患者 0.5 g 左右新鲜粪便于培养瓶,使用 1:100 法稀释至 10-8,滴种于相应培养基,主要滴种菌群为双歧杆菌、乳杆菌、酵母菌、梭菌;结果以常用对数值(Ig 菌落数 /g)表示。

**1.3.5 不良反应的发生情况。**

### 1.4 统计学分析

以 spss18.0 软件包处理实验数据,计量资料用均数± 标准差($\bar{x}$± s)表示,组间比较采用 t 检验,计数资料组间比较采用 $x^2$ 检验,以 P<0.05 表示差异具有统计学意义。

## 2 结果

### 2.1 两组治疗前后临床症状的比较

治疗前,两组腹痛、腹泻、脓血便评分比较差异无统计学意义(P>0.05);治疗后,两组腹痛、腹泻、脓血便评分均较治疗前均显著降低(P<0.05),且观察组腹痛、腹泻、脓血便评分均明显低于对照组(P<0.05),见表 1。

**表 1 两组治疗前后临床症状比较($\bar{x}$± s,分)**

Table 1 Comparison of the clinical symptom before and after treatment between two groups($\bar{x}$± s, scores)

| Groups | | Bellyache | Diarrhea | Pus and blood stool |
|---|---|---|---|---|
| Observation group(n=46) | Before treatment | 3.87± 0.32 | 3.64± 0.35 | 3.39± 0.34 |
| | After treatment | 1.43± 0.26*# | 1.24± 0.28*# | 1.06± 0.13*# |
| Control group(n=46) | Before treatment | 3.90± 0.31 | 3.61± 0.40 | 3.42± 0.30 |
| | After treatment | 2.25± 0.27* | 2.27± 0.33* | 2.07± 0.16* |

Note: Compared with the before treatment, *P<0.05; Compared with the control group, #P<0.05.

### 2.2 两组治疗前后 Baron 内镜评分的比较

治疗前,两组 Baron 内镜评分比较差异无统计学意义(P>0.05);治疗后,两组 Baron 内镜评分均较治疗前明显降低(P<0.05),观察组 Baron 内镜评分明显低于对照组(P<0.05),见表 2。

### 2.3 两组治疗前后肠粘膜 TLR4、NF-κb 表达的比较

治疗前,两组肠粘膜 TLR4、NF-κb 表达比较差异无统计学意义(P>0.05);治疗后,两组肠粘膜 TLR4、NF-κb 表达均较治疗前明显降低(P<0.05),且观察组肠粘膜 TLR4、NF-κb 表达均明显低于对照组(P<0.05),见表 3。

### 2.4 两组治疗前后肠道菌群数量的比较

治疗前, 两组各肠道菌群数量比较差异均无统计学意义(P>0.05);治疗后,两组双歧杆菌、乳杆菌、酵母菌、梭菌数量均较治疗前均显著改变(P<0.05),且观察组双歧杆菌、乳杆菌数量明显多于对照组, 酵母菌、梭菌数量明显少于对照组 (P<

0.05),见表 4。

表 2 两组治疗前后 Baron 内镜评分的比较($\bar{x}\pm s$,分)

Table 2 Comparison of the Baron endoscopic score before and after treatment between two groups($\bar{x}\pm s$, scores)

| Groups | | Baron endoscopic scores |
|---|---|---|
| Observation group(n=46) | Before treatment | 2.42± 0.27 |
| | After treatment | 1.13± 0.16*# |
| Control group(n=46) | Before treatment | 2.47± 0.25 |
| | After treatment | 1.82± 0.19* |

Note: Compared with the before treatment, *P＜0.05; Compared with the control group, #P＜0.05.

表 3 两组治疗前后肠粘膜 TLR4、NF-κb 表达的比较($\bar{x}\pm s$,OD 值)

Table 3 Comparison of the TLR4 and NF-κb expressions in the intestinal mucosa before and after treatment between two groups($\bar{x}\pm s$, OD value)

| Groups | | TLR4 | NF-κb |
|---|---|---|---|
| Observation group(n=46) | Before treatment | 0.39± 0.05 | 0.41± 0.06 |
| | After treatment | 0.17± 0.02*# | 0.18± 0.03*# |
| Control group(n=46) | Before treatment | 0.42± 0.04 | 0.39± 0.07 |
| | After treatment | 0.29± 0.03* | 0.30± 0.04* |

Note: Compared with the before treatment, *P＜0.05; Compared with the control group, #P＜0.05.

表 4 两组治疗前后肠道菌群数量的比较($\bar{x}\pm s$,Ig)

Table 4 Comparison of the intestinal flora before and after treatment between two groups($\bar{x}\pm s$, Ig)

| Groups | | *Bacillus bifidus* | *Lactobacilli* | *Microzyme* | *Clostridium* |
|---|---|---|---|---|---|
| Observation group(n=46) | Before treatment | 9.53± 0.62 | 9.83± 0.58 | 11.25± 0.62 | 11.74± 0.67 |
| | After treatment | 11.75± 0.83*# | 11.48± 0.74*# | 9.48± 0.43*# | 9.59± 0.40*# |
| Control group(n=46) | Before treatment | 9.57± 0.60 | 9.79± 0.62 | 11.28± 0.60 | 11.69± 0.70 |
| | After treatment | 10.48± 0.70* | 10.62± 0.67* | 10.39± 0.52* | 10.73± 0.62* |

Note: Compared with the before treatment, *P＜0.05; Compared with the control group, #P＜0.05.

### 2.5 两组不良反应发生情况的比较

两组治疗过程中均未有明显不良反应出现,治疗前后肝肾功能、血常规等比较差异均无显著统计学意义(P＞0.05)。

## 3 讨论

目前,溃疡性结肠炎的发病机制仍不完全明确,研究认为与遗传、免疫紊乱、肠道感染、环境因素等存在着密切的关系[10]。该病的治疗尚未有特殊疗法,主要给予抗炎药物的口服治疗。美沙拉秦主要成分为 5- 氨基水杨酸,可通过对易引发结肠黏膜炎症介质产生抑制作用,例如前列腺素 E2、白三烯 B4、血小板活动因子等,缓解肠道炎症,但在停药后易复发[11,12]。

益生菌作为对宿主有益的活性微生物,主要在肠道、生殖系统内定值,有助于对宿主微生态平衡进行改善。随着益生菌应用范围的不断扩增, 目前也已逐渐应用到炎症性肠病中,其中常用的益生菌有乳酸杆菌、双歧杆菌、酵母菌、衣地芽胞杆菌、乳酸球菌等。思连康是种双歧杆菌四联活菌,包含双歧杆菌、乳杆菌、肠球菌以及蜡样芽孢杆菌,可直接补充机体正常生理细菌,在肠道形成一个生物屏障,继而促进肠道蠕动,缓解临床症状[13,14]。Yasueda A 等[15]认为益生菌不仅可达到与抗炎药物相似的治疗效果,且可对机体局部、系统的免疫反应进行调节。本研究结果显示联合用药的患者治疗后腹痛、腹泻、脓血便以及 Baron 内镜评分均明显比单独用药的患者低,显示出联合用药在缓解临床症状上更具有优势,和 Tamaki H 等[16]研究具有相似性。此外,在治疗过程中均未有严重不良反应发生,血常规、肝肾功能无明显改变,提示思连康用于溃疡性结肠炎的治疗中安全性高。

近年来,Toll 样受体(TLR)家族在天然免疫反应的启动作用已备受学者关注, 通过识别和结合病原识别模式分子活化 MyD88-IRAK-NF-κb 通路, 令 NF-κb 的核转位结构域得以暴露,继而 NF-κb 进入核内,并对各类炎症因子的转录产生激活作用。而不同的 TLR 所识别的配体不同,Atreya R 等[17]报道显示在肠道粘膜中,TLR4 识别革兰阴性杆菌的脂多糖,在炎症反应中作用关键。Fernandes P 等[18]的进一步研究中也显示在溃疡性结肠炎患者的肠粘膜中,TLR-4 以及其下游信号分子 NF-κb 的表达明显较正常人群高。此外,肠道菌群失调在溃疡性结肠炎患者的发生也存在着密切关系,其机制主要为肠道菌群紊乱所造成的肠粘膜发生过度免疫反应,破坏肠粘膜完整性,损伤到正常的防御功能,从而诱发疾病。国内外均有学者证实溃疡性结肠炎患者具有不同程度的肠道菌群失调表现,例如益生菌(双歧杆菌、乳杆菌等)数量明显减少,致病菌(例如酵母菌、梭菌)数量明显增加等;益生菌数量的减少,会削弱其和上皮表面特异性抗体所形成的菌膜屏障,令肠道定值抗力降低,增加致

病菌对肠粘膜上皮细胞的破坏和侵袭,引发肠道慢性炎症[19,20]。Gallo A 等[21]发现在溃疡性结肠炎患者中,肠道炎症、菌群紊乱等因素损伤肠道粘膜功能,促炎因子等侵入粘膜固有层,继而增加肠道炎症,可形成一种恶性循环。本研究结果显示两组患者在治疗后肠粘膜 TLR4、NF-κb 的表达均出现降低,但联合用药的患者降低程度更明显,且双歧杆菌、乳杆菌明显增加,酵母菌、梭菌明显减少,效果均优于单独使用美沙拉秦的患者,显示出联合用药发挥相互协同作用,在抑制肠粘膜中 MyD88-IRAK-NF-κb 通路的激活中效果更为显著[22]。Yoshimatsu Y 等[23]研究也证实在常规抗炎药物基础上联用益生菌治疗不仅可有效降低肠粘膜炎症反应,且可进一步恢复肠道菌群的平衡,这也可能是联合用药患者临床症状改善更明显的内在机制之一。但本研究随访时间较短,对于远期复发率上仍需进一步深入研究。

综上所述,在溃疡性结肠炎患者中应用思连康联合美沙拉秦效果显著,可有效缓解临床症状,促进粘膜修复,其内在机制可能和降低肠粘膜 TLR4、NF-κb 的表达及调节肠道微生态平衡相关。

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