MONITORING SULFASALAZINE THERAPY IN PATIENTS WITH CHRONIC INFLAMMATORY BOWEL DISEASE

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ABSTRACT

Sulfasalazine consists of sulfapyridine and 5-aminosalicylic acid moieties linked by an azo bond. It is used in the treatment of active inflammatory bowel disease and is the most effective agent for maintaining remission in chronic inflammatory bowel disease. A study conducted by Das and co-workers (Das et al, 1973b) indicated the therapeutic efficacy of sulfasalazine was related to serum concentrations of total sulfapyridine metabolites above 20 µg/ml. This investigation was expanded to study the relationship between adverse effects associated with sulfasalazine therapy and serum levels of sulfapyridine metabolites in a larger number (133) of patients (Das et al, 1973c). The occurrence of toxicity appeared to be related to serum conentrations of total sulfapyridine metabolites above 50 µg/ml. This and subsequent studies have shown an increased incidence of toxicity among patients who are slow acetylators of the sulfapyridine moiety.

To obtain more information about the relationships between serum concentrations of sulfapyridine metabolites, acetylator phenotype, and efficacy and toxicity of sulfasalazine therapy, a sensitive analytical procedure was needed. Therefore, a rapid, specific and sensitive high pressure liquid chromatographic procedure was developed for determination of sulfapyridine (SP) and its major metabolite, N^4-acetylsulfapyridine (AcSP), in biological fluids. Calibration curves were linear and the mean recovery of SP (AcSP) from serum, saliva and urine was 91.5 (82.0),

88.6 (72.6) and 98.6 (90.6) %, respectively. A sensitive and specific gas chromatographic procedure for determination of SP and AcSP was also developed. However, this procedure was cumbersome in comparison with the high pressure liquid chromatographic procedure and was not used for monitoring sulfasalazine metabolites in the clinical study. The traditional spectrophotometric assay lacked specificity and did not provide adequate sensitivity for determination of sulfasalazine metabolites in small volumes of biological fluids.

A clinical study was conducted in 28 outpatients with active or quiescent chronic inflammatory bowel disease. Concentrations of SP and AcSP were determined in samples of serum, saliva and urine (24 h urine collection or a single collection) obtained from each patient. The acetylator phenotype ('rapid' or 'slow') of each patient was assessed according to the percentage of acetylated SP (% acetylation) in serum. Calculation of the % acetylation in serum provided a clear distinction between rapid and slow acetylators. In comparison, assessment of acetylator phenotype according to % acetylation in saliva or urine was less accurate.

Serum concentrations of SP ranged from 3.5 to 73.1 µg/ml for patients with controlled disease and 6.3 to 38.0 µg/ml for patients with active disease ; serum concentrations of total SP (SP + AcSP), from 5.7 to 95.1 µg/ml for patients with controlled disease and 14.0 to 54.7 µg/ml for patients with active disease. No correlation between clinical status and serum levels of SP or total SP was detected using

multiple linear regression analysis ($p > 0.05$). However, when the data were analyzed separately for rapid and slow acetylators, in rapid acetylators controlled disease was associated with elevated serum SP concentrations ($p > 0.05$). For slow acetylators clinical status made no significant contribution to regression ($p > 0.05$).

In contrast to previous studies (Das et al, 1973b; Goldstein et al, 1978) sulfasalazine dosage, expressed in mg/kg of body weight, was found to be an important determinant of serum concentrations of SP and total SP for both slow and rapid acetylators. A positive correlation between the percentage acetylation in serum and the concentration of unmetabolized SP in serum was found in the 28 patients studied. However, when the data were analyzed separately for rapid and slow acetylators, there was no correlation between percentage acetylation and serum concentrations of unmetabolized SP (multiple linear regression analysis, $p > 0.05$ in both cases).

Slow acetylators had significantly higher serum concentrations of SP (21.9 µg/ml) than rapid acetylators (8.8 µg/ml) (oneway analysis of variance, $p < 0.05$). The incidence of toxicity was higher in slow acetylators but did not reach statistical significance (chi-square test, $p > 0.05$), probably because of the small number of patients studied. Serum concentrations of SP were significantly higher in patients who reported toxic effects (23.2 µg/ml) than in patients who did not report toxicity (13.9 µg/ml) (oneway analysis of variance, $p < 0.05$). However, serum concentrations of total SP (SP + AcSP) were not significantly

different among patients with (32.9 µg/ml) or without (22.7 µg/ml) toxic effects (oneway analysis of variance, $p > 0.05$).

To determine whether drug concentrations in saliva correlate with those in serum, the saliva/serum ratio for SP and AcSP was calculated for each patient. The mean concentrations ratio (± S.D.) for SP and AcSP was 0.55 (± 0.09) and 0.18 (± 0.12), respectively. Because of the high inter-individual variability in the distribution of SP and AcSP in saliva, determination of saliva concentrations did not provide a reliable estimate of serum levels.

For clinical evaluation of inflammatory bowel disease patients determination of serum SP concentrations appears to be more important for monitoring toxicity than therapeutic efficacy of sulfasalazine. Assessment of acetylator status appears to be useful for predicting serum SP levels in patients receiving sulfasalazine therapy. Determination of drug concentrations in saliva does not appear to be a useful method for estimating serum drug levels or assessing acetylator status because of the variable distribution of SP and AcSP in saliva.

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INTRODUCTION

Ulcerative colitis and Crohn's disease are the primary forms of chronic inflammatory bowel disease. Both diseases are characterized by their chronic nature and significant morbidity in most patients. Because the etiology of these disorders has remained obscure, therapy has been mainly symptomatic. Treatment, directed at controlling the symptoms and improving the quality of life, is usually prolonged. Accepted treatment includes the use of sulfasalazine and steroids. While sulfasalazine remains the mainstay of therapy in quiescent disease, steroids and supportive measures are added in moderate to severe disease.

Although the efficacy of sulfasalazine in inflammatory bowel disease is well documented, the mechanism of action remains unknown. Clinical improvement and adverse effects seem to correlate with serum concentrations of certain of sulfasalazine's metabolites. The purpose of this study was to monitor the disposition of the sulfasalazine metabolites, sulfapyridine and N^4-acetylsulfapyridine, in chronic inflammatory bowel disease patients and to investigate a possible relationship with toxicity and/or efficacy.

I. Chronic Inflammatory Bowel Disease

Ulcerative colitis (UC) and Crohn's disease (CD) are both described as chronic inflammatory bowel disease (CIBD). CD is also referred to as regional enteritis, regional enterocolitis, granulomatous colitis, and transmural colitis. Terms such as CD of the ileum, colon and jejunum designate the area of the bowel affected (Poley, 1978a; Smith and Tong, 1979).

Although CD and UC exhibit similar clinical, epidemiologic, and pathologic features, in 80 to 90% of cases of CIBD they can be distinguished as separate clinical entities (Glickman and Isselbacher, 1980; LaMont and Isselbacher, 1980). CD and UC are found more frequently in Caucasians than non-Caucasians, in urban than rural settings, and in Jews than non-Jews (Smith and Tong, 1979; Poley, 1978a; LaMont and Isselbacher, 1980). In the western world, the incidence of UC (including ulcerative proctitis) is under 10 per 100,000 white adults at risk, CD of the colon, 2 to 3 per 100,000, and CD of the small intestine, 2 per 100,000. The incidence of both UC and CD appears to be increasing.

UC and CD commonly affect adolescents and young adults. Although CIBD is found in all age groups, usually the disease is initially detected in the first to fourth decade of life (Poley, 1978a; Smith and Tong, 1979). The incidence of CIBD in males and females is similar.

The etiology of CIBD is unknown. Familial clustering and association

with diseases that have a hereditary basis suggest that UC and CD are closely related (LaMont and Isselbacher, 1980). UC and CD may represent variations in the host response to the same etiologic agent. Although multiple etiologic factors seem to be involved, their relationship to the clinical course, symptoms, and response to treatment remains unresolved.

UC and CD are characterized by an inflammation of the mucosal epithelial cells (and submucosa in the case of CD). (Poley, 1978a; Glickman and Isselbacher, 1980; LaMont and Isselbacher, 1980). The clinical manifestations of this inflammatory process reflect the location, extent, and severity of the lesion. The proctoscopic, radiologic, sigmoidoscopic, and clinical findings for UC and CD are summarized and compared in Table I. A variety of systemic and extracolonic complications can occur as a consequence of CIBD (Table I).

In addition to UC, CD of the colon, and CD confined to the small intestine, ulcerative proctitis is included within the general classification of chronic inflammatory bowel disease (LaMont and Isselbacher, 1980). Ulcerative proctitis is a mild form of UC which often presents as rectal bleeding and constipation (rather than diarrhea). Sigmoidoscopy reveals a friable mucosa and an inflammatory exudate. Except in long-standing disease the bowel usually appears normal on radiologic examination. The incidence of local and systemic complications, including

Table I: Characteristics of Chronic Inflammatory Bowel Disease

	Ulcerative Colitis	Crohn's Disease
Etiology		
Transmissible agent, possibly a virus	+	+
Autoimmune component, possibly an abnormality in cell-mediated immunity	+	+
Underlying allergy or hypersensitivity	+	+
Psychological features	+	+
Infectious basis	-	+
Pathophysiology		
Depth of involvement	Mucosa and submucosa; hyperemia and ulcer formation	Transmural, leading to thickening of the bowel and narrowing of the lumen.
Granulomas	Rare	Common; present in 50% of cases; a distinguishing feature.
Fissures, fistulas	Rare	Common; a result of penetrating inflammation and ulceration
Radiologic Findings		
Type of involvement	Continuous	Discontinuous
Colonic involvement	Predominately left-sided and/or distal	60% of patients have colonic involvement, 10% colon-only; frequent right colon involvement
Terminal ileum	Widened, patulous if involved; the remainder of the small bowel is not involved	Most common location of the disease; narrow, stiff ileum. The small bowel is often involved

Sigmoidoscopic Findings		
Rectal involvement	95%	50%
Friable mucosa	Frequent	Uncommon
Clinical Findings		
Rectal bleeding	Common, continuous	Infrequent
Diarrhea/other symptoms	Frequent history of diarrhea of long duration (semi-formed to liquid, bloody, purulent stools, abdominal pain, weight loss and fever	Frequent history of abdominal fever, weight loss, and diarrhea
Perianal, perirectal symptoms	Occasional abscess	Greater propensity to fistulation; perianal disease is common; fissures and ulcers more common with colonic involvement
Toxic megacolon	Occasional, part of the spectrum of fulminating disease	Rare; part of the spectrum of fulminating disease
Incidence of carcinoma	Increased risk when the disease develops in childhood	Rare. Increased risk compared with the general population
Systemic Complications		
Nutritional and metabolic changes	Muscle wasting, electrolyte losses, negative nitrogen balance, hypoalbuminemia, iron-deficiency anemia, vitamin deficiences, and growth retardation in children (more common with CD)	
Arthritis	Common	Common

Systemic Complications-cont'd.		
Liver disease	Common	Common, severe lesions are found less frequently than with UC
Skin disorders	Common	Common
Uveitis	Common	Common
Stomatitis, peptic ulcer disease	Occasional	Common
Venous thrombosis and thromboembolism	Common	Common
Renal Stones	Occasional	Occasional, in the presence of ileal disease
Prognosis	Varies with severity and age; less favorable in patients with extensive involvement and moderate activity, and when disease activity begins in childhood. Following medical treatment, remission rate is 90% for mild attacks and 50% for severe attacks; 75% will experience relapse which carries a worse prognosis. For the initial attack, 20 to 25% will require colectomy.	Varies with severity; generally follows a chronic, intermittent course usually marked by one or more complications. In comparison with UC it has a more unrelenting progression, relative refractoriness to medical treatment, and a greater tendancy to postoperative recurrence.
Mortality	In patients with moderate to moderately severe attacks, the 5-year mortality rate ranges from 5 to 15%	In patients with extensive disease accompanied by complications and frequent hospitalizations, there is a 5 to 10% mortality rate regardless of treatment.

carcinoma, is reduced in comparison with UC.

The medical management of CIBD is largely non-specific and non-curative and is hampered by the unpredictable, often explosive, clinical course (Poley, 1978b; Green, 1979; LaMont and Isselbacher, 1980).

A rational therapeutic regimen can be designed once patients have been classified according to severity of the disease (Table II lists clinical criteria which are considered helpful in judging the severity of the disease) (Poley, 1978a, 1978b; Green, 1979). The initial investigation is limited to those procedures and laboratory tests required to make a diagnosis and to distinguish patients with moderate to severe symptoms. An acute attack of moderate to severe CIBD is best treated in a hospital setting where many patients will respond to bed rest, a clear liquid diet, and intravenous feedings to replace fluid and electrolyte loss (Green, 1979; LaMont and Isselbacher, 1980). Opiates or diphenoxylate may be used in the treatment of diarrhea and abdominal cramps only in the absence of intestinal obstruction because of the increased risk of toxic megacolon associated with these drugs.

Re-establishing a positive nutritional balance to aid healing of the ulcerative process and restore body weight is an important aspect of the medical management of CIBD (LaMont and Isselbacher, 1980; Poley, 1978b; Green, 1979). Where this cannot be achieved by oral nutrition, as in severely ill patients with extensive intestinal involvement, intravenous alimentation has been successfully employed. Total

Table II: Criteria for Assessment of Disease Activity in Ulcerative Colitis and Crohn's Disease

Finding	Mild[a]	Moderate to Severe
Ulcerative Colitis		
Diarrhea	Less than 4-5 stools per day	Greater than 6-10 loose stools per day
Fever	Minimal	Greater than 38°
Blood loss/Anemia	Minimal blood loss	Blood loss sufficient to establish an anemia of Hb < 11.5 gm/dl (moderate) or Hb < 10 gm/dl (severe)
Pain	Minimal	Severe abdominal pain
Tachycardia	Absent	Greater than 100 beats/min
Leukocytosis	Absent	Greater than 10,000/mm^3
Erythrocyte sedimentation rate	Normal	Greater than 30 mm/h
Extracolonic Manifestations	Absent	Present
Crohn's Disease		
Diarrhea	1-4 stools per day	Greater than 6 stools per day
Weight loss	Less than 10 kg	Greater than 10 kg
Pain	Minimal	Severe abdominal pain
General condition	Good	Poor, malnutrition may be apparent
Anemia	Mild or absent	Anemia of Hb < 11.5 gm/dl
Erythrocyte sedimentation rate	Moderately elevated or normal	Greater than 30 mm/h
Serum albumin	Normal or slightly decreased	Less than 3.0 gm/dl
Thrombocytosis	Slight or absent	Greater than 400,000/mm^3

[a] In children, mild disease activity is characterized by few stools with some blood and mucus.

(Adapted from Poley, 1978a, Table 3, p. 940)

parenteral nutrition and complete bowel rest has successfully controlled the disease process, eliminated the need for surgical intervention, and reversed growth arrest in children. Elemental or low residue diets also provide adequate nutrition and low fecal volume. The bowel is allowed to rest and the bacterial population to concommitantly decrease.

Sulfasalazine is used widely to treat CIBD in adults and children. The major goal of sulfasalazine therapy is to prevent recurrences of UC and CD of the colon (Poley 1978b; Smith and Tong, 1979; Green, 1979; Northfield, 1979, LaMont and Isselbacher, 1980). The mechanism of sulfasalazine's beneficial effect is unknown. Some authors classify sulfasalazine as an anti-inflammatory drug while others retain the traditional antimicrobial classification.

When supportive therapy fails to provide clinical improvement in CIBD steroid therapy is often effective, especially in patients with ileal involvement (Summers et al, 1979; Glickman and Isselbacher, 1980; LaMont and Isselbacher, 1980). The beneficial effects of corticosteroids in ulcerative colitis may be derived from their anti-inflammatory activity and/or their immunosuppressive properties. Adrenocorticotrophic hormone (ACTH), hydrocortisone, prednisone, and prednisolone are the drugs of choice for inducing remission in the acute stages of moderate to severe UC and CD. If oral therapy is chosen, usually prednisone is initiated at a dose of 40 to 60 mg per day and tapered gradually as improvement is noted (usually after 7 to 10 days).

Some patients may require a daily maintenance dose of 10 to 15 mg of prednisone in combination with sulfasalazine to control symptoms. Administration of corticosteroids on alternate days may reduce side effects but may be less efficacious. Steroids should not be used as maintenance therapy in quiescent disease as they have no value in preventing recurrences.

Topical use of steroids has been recommended for patients with ulcerative proctitis (LaMont and Isselbacher, 1980; Serebro et al, 1977; Moller et al 1978). The usual regimen is 20 mg of prednisone or 100 mg of hydrocortisone administered rectally (by retention enema) at bedtime. Steroid enemas carry a minimal risk of side effects as a result of systemic absorption.

Azathioprine has been the most frequently used immunosuppresive drug for treatment of CIBD (Glickman and Isselbacher, 1980; LaMont and Isselbacher, 1980; Poley, 1978b). Azathioprine is only useful in maintenance of remission and is generally reserved for patients who do not tolerate steroids or sulfasalazine. Other agents which have been used in inflammatory bowel disease include 6-mercaptopurine, disodium cromoglycate, metronidazole, and immunotherapy (transfer factor, levamisole) (Green, 1979).

In patients with CIBD there appears to be a complex interplay between psychological responses and disease activity. Most therapists

agree that routine psychotherapy is unnecessary for CIBD patients, although supportive psychotherapy in the form of a strong physician-patient relationship is extremely important (Poley, 1978b; LaMont and Isselbacher, 1980). In certain instances formal psychiatric assistance is necessary.

Surgical therapy for CD is generally reserved for complications rather than primary treatment but may be indicated in severe, intractable UC. Indications for surgical therapy include toxic megacolon, stricture, suspected malignancy, massive hemorrhage and perforation (Poley, 1978b; Green, 1979; LaMont and Isselbacher, 1980). Surgical intervention may be used more frequently in pediatric patients to restore growth and development and to prevent carcinoma. Approximately 80% of patients with CD will require surgery within 20 years of the onset of disease (Hagop et al, 1979). Drug treatment does not alter the need for surgical intervention and recent studies suggest that it may not affect the recurrence rate of CD (Summers et al, 1979). CIBD may cause disability in every aspect of the patient's life (Mallett et al, 1978). The most frequent causes of limitation are bowel frequency, urgency of defecation, abdominal or rectal pain and lassitude. However, through meticulous supportive and nutritional therapy CIBD patients are able to function reasonably well.

II. Sulfasalazine

A. History

Sulfasalazine (salicylazosulfapyridine, SASP) (Figure 1 (a)) consists of a sulfapyridine moiety (Figure 1 (b)) linked by an azo bond to 5-aminosalicylic acid (Figure 1 (c)). SASP was introduced in Sweden in 1939 by Dr. Nanna Svartz as the most promising of a number of salicylic - sulfonamide combinations developed for the treatment of polyarthritis (Svartz, 1942). SASP is now used in the treatment of inflammatory bowel disease. The popularity of SASP is based on early clinical trials which demonstrated a favorable response to the drug in the majority of patients (Collins, 1968). Svartz introduced the use of SASP in treatment of UC in 1942. However, a controlled trial of SASP therapy in outpatients was not conducted until 20 years later (Baron et al, 1962). The relative efficacy of SASP in short term therapy of an acute attack of mild UC was compared with salicylazosulfadimidine and a placebo. SASP produced remission in 25% of cases while salicylazosulfadimidine was no better than the placebo. At the same time a similar short term trial established the superiority of combined oral and topical steroid therapy over SASP for acute attacks of UC (Truelove et al, 1962). In controlled trials conducted in 1965 SASP was superior to a placebo in decreasing the rate of relapse in patients in remission from proctocolitis (Misiewicz et al, 1965), while corticosteroids were ineffective in preventing relapses of ulcerative

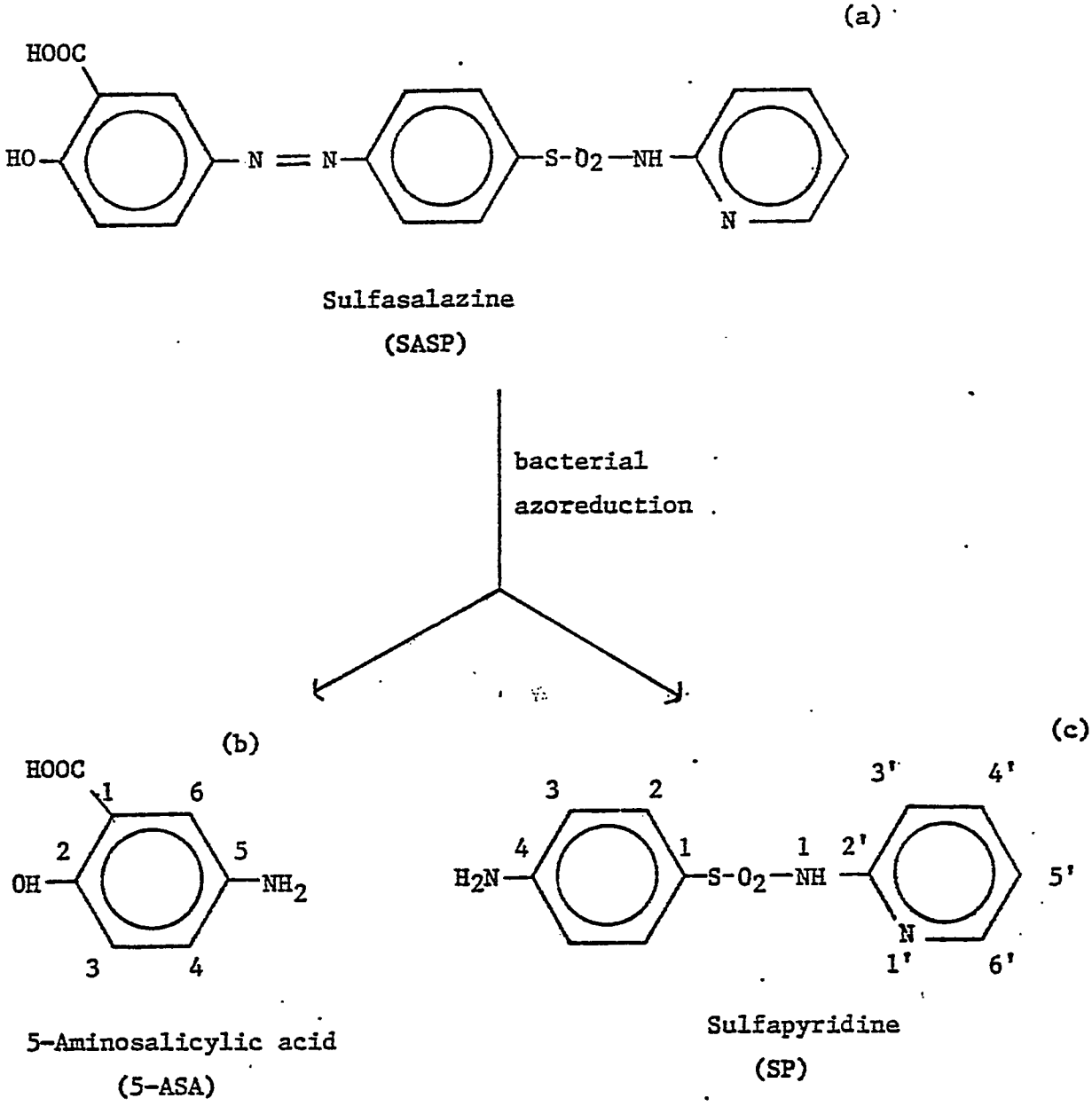

Figure 1 : Colonic metabolism of sulfasalazine in man.

colitis (Lennard-Jones et al, 1965).

B. Therapeutic Use

Sulfasalazine is used widely in the treatment of UC and CD. In combination with the usual dietary measures, SASP is frequently used in mild to moderate UC (Das and Dubin, 1976). The recommended initial dose of SASP in patients with mild UC or CD is 0.5 to 1.0 g four times daily with meals (LaMont and Isselbacher, 1980; Weinstein, 1975). A daily dose of 2 to 3 g may be administered as an enema to induce remission in ulcerative proctitis (Khan and Piris, 1977; Moller et al, 1978) or for initial and maintenance therapy of UC affecting the rectum and descending colon (Serebro et al, 1977).

The drugs used for initial treatment of moderate to severe UC are corticosteroids or ACTH (LaMont and Isselbacher, 1980). Following a reduction in inflammation, steroid therapy is usually tapered and SASP therapy is instituted at 0.5 g twice daily and increased over several days to 0.5 g four times daily. Unless symptoms persist or worsen, steroids are continually tapered and then withdrawn completely to avoid their long-term side-effects. A maintenance dose of 2 g SASP per day is usually sufficient and should not be increased beyond 6 g daily (LaMont and Isselbacher, 1980; Goldstein et al, 1979; Poley, 1978b; Smith and Tong, 1979). There is no evidence that higher doses of SASP provide additional benefit.

In children a daily SASP dose of 50 to 100 mg/kg is adequate at the

onset of therapy. Daily doses of 40 to 70 mg/kg usually produce safe and effective blood levels (Poley, 1978b; Goldstein et al, 1979).

Both SASP and prednisone are effective when given alone for active CD and are widely used in combination (Singleton et al, 1979b). A recent study (Trial of Adjunctive Sulfasalazine in Crohn's Disease) compared the effect of SASP in combination with prednisone with that of prednisone and placebo in patients with active CD. Sixty percent of patients achieved remission during treatment with prednisone-placebo, but SASP appeared to slow and blunt the response to prednisone. Therefore, SASP and prednisone should not be used in combination for active CD. Furthermore, in patients with quiescent disease, SASP did not appear to exert a prednisone-sparing effect (i.e., allowing a more rapid or complete withdrawal of prednisone from patients who have responded to it).

According to the National Cooperative Crohn's Disease Study (NCCDS), SASP (1 g per 15 kg) is effective mainly in patients with CD of the colon (Summers et al, 1979). Controlled trials of SASP therapy in CD have been conducted only recently (Wenkert et al, 1978; Summers et al, 1979). Nevertheless, SASP has been widely used in treatment of CD probably because of its efficacy in UC. Patients with CD of the ileum and colon will benefit from either SASP or prednisone, whereas only prednisone is effective in patients with only small bowel involvement. If patients do not respond to prednisone they are unlikely to benefit

from substitution of SASP. SASP is the drug of choice for initial therapy and has been proven to be the safest of the drugs used to treat CD. Results of the NCCDS also indicated that therapy with SASP (0.5 g per 15 kg), prednisone (0.25 mg per kg) or azathioprine (1 mg per kg) will not exert a prophylactic effect and may produce more problems than no therapy in patients with quiescent disease or after extirpative therapy (Summers et al, 1979). According to the Inter-Nordic Cooperative Study on SASP in CD, SASP may be effective as prophylaxis in patients who have not undergone extirpative surgery (Wenkert et al, 1978). However, Summers et al (1979) concluded that no drug should be used prophylactically in patients with quiescent disease or after extirpative surgery.

C. Pharmacokinetics of sulfasalazine and its metabolic products

1. Sulfasalazine

Approximately one-quarter to one-third of the dose of SASP administered orally is absorbed intact from the small bowel (Schroder and Campbell, 1972; Das et al, 1979). Peak blood levels are reached four to five hours after oral administration (Schroder and Campbell, 1972). SASP is not metabolized by the liver and is excreted unchanged in the bile (Das et al, 1979). A small fraction of the dose enters the circulation and is excreted in the urine (less than 10 percent of the dose may be recovered in the urine as unchanged SASP) (Schroder and