

**NON-STEROIDAL
ANTI-INFLAMMATORY DRUGS
AND PEPTIC ULCERS**

Ali S A Taha MD (Glasgow), MRCP (UK)

Thesis submitted for the degree of Doctor of Philosophy
to the University of Glasgow

from the
Gastroenterology Unit
Royal Infirmary
Glasgow UK

1993

ProQuest Number: 13833429

All rights reserved

INFORMATION TO ALL USERS

The quality of this reproduction is dependent upon the quality of the copy submitted.

In the unlikely event that the author did not send a complete manuscript and there are missing pages, these will be noted. Also, if material had to be removed, a note will indicate the deletion.



ProQuest 13833429

Published by ProQuest LLC (2019). Copyright of the Dissertation is held by the Author.

All rights reserved.

This work is protected against unauthorized copying under Title 17, United States Code
Microform Edition © ProQuest LLC.

ProQuest LLC.
789 East Eisenhower Parkway
P.O. Box 1346
Ann Arbor, MI 48106 – 1346

Thesis
9670
copy 1

GLASGOW
UNIVERSITY
LIBRARY

PREFACE

The studies contained in this Thesis were conducted during my appointment at the Gastroenterology Unit, Royal Infirmary, Glasgow; some of them have already been published and the rest are being considered for publication. The majority of these studies have also been presented to learned societies in and outside the UK. The contribution of my co-workers is formally acknowledged, and the work and writing of this Thesis has been personally carried out by me.

CONTENTS OF THESIS

Contents of Thesis

	PAGE
Preface	2
Content	3
Acknowledgements	11
Summary	14
Section 1	
Background of Thesis	22
Introduction	23
Chapter 1	
Mechanisms of NSAID - related peptic damage and their implications for management	24
1.1	
Introduction	24
1.2	
NSAID and mucosal prostaglandins	24
1.3	
Other mechanisms of NSAID - related mucosal toxicity	26
1.4	
Implications for management	28

	PAGE
Chapter 2 The Interaction between NSAID and <i>Helicobacter pylori</i>	33
2.1 Summary	33
2.2 Introduction	34
2.3 Effect of NSAID and <i>H pylori</i> on gastric acid secretion	34
2.4 The mucus layer	35
2.5 Mucosal prostaglandins	36
2.6 Mucosal blood flow	37
2.7 Histological and endoscopic abnormalities	38
2.8 Therapeutic considerations	41
Chapter 3 NSAID - related ulcers and anti-rheumatoid second-line drugs	43
Section II Aims of Thesis	48
Section III Diagnostic studies	52
Introduction	53

	PAGE	
Chapter 4	Influence of NSAID on the efficacy of diagnostic test for H pylori	54
4.1	Summary	54
4.2	Introduction	55
4.3	Patients and methods	55
4.4	Results	58
4.5	Discussion	67
Chapter 5	Effects of NSAID on the sero-diagnosis of H pylori	70
5.1	Summary	70
5.2	Introduction	71
5.3	Patients and methods	72
5.4	Results	75
5.5	Discussion	81
Chapter 6	Prediction of NSAID - related peptic ulcers using the Health Assessment Questionnaire	86

	PAGE
6.1 Summary	86
6.2 Introduction	87
6.3 Patients and methods	87
6.4 Results	89
6.5 Discussion	96
Section IV NSAID - related histological changes in the upper gastrointestinal tract	98
Introduction	99
Chapter 7 NSAID and the gastric mucosa	100
7.1 Summary	100
7.2 Introduction	101
7.3 Patients and methods	102
7.4 Results	107
7.5 Discussion	115

		PAGE
Chapter 8	NSAID and the duodenal mucosa	120
8.1	Summary	120
8.2	Introduction	121
8.3	Patients and methods	122
8.4	Results	125
8.5	Discussion	134
Chapter 9	NSAID and the oesophagus	138
9.1	Summary	138
9.2	Introduction	139
9.3	Patients and methods	140
9.4	Results	144
9.5	Discussion	155
Section V	NSAID and mucosal blood flow	160
Chapter 10	Gastric and duodenal blood flow in patients receiving NSAID: influence of age, smoking, ulceration, and H pylori	161

	PAGE
10.1 Summary	161
10.2 Introduction	162
10.3 Patients and methods	163
10.4 Results	165
10.5 Discussion	171
Section VI NSAID - related ulcers and second-line drugs	175
Introduction	176
Chapter 11 Endoscopic non-steroidal peptic damage in rheumatoid patients receiving second-line drugs	177
11.1 Summary	177
11.2 Introduction	178
11.3 Patients and methods	178
11.4 Results	179
11.5 Discussion	185

Chapter 12	NSAID - related ulcers and H pylori in rheumatoid arthritis patients receiving gold or sulphasalazine	188
12.1	Summary	188
12.2	Introduction	189
12.3	Patients and methods	190
12.4	Results	191
12.5	Discussion	197
Section VII	Conclusions	201
	References	206
Section VIII	Publications related to this Thesis	240
APPENDIX		247

ACKNOWLEDGEMENTS

Acknowledgements

The help, advice, and active participation of a number of friends and colleagues in various units and departments at Glasgow Royal Infirmary, have all been vital for the completion of studies contained in this Thesis.

I would like to start by thanking Dr R I Russell, Head of the Gastroenterology Unit, for his invaluable advice and supervision. Dr Russell was kind enough to make available the various facilities at the Gastroenterology Unit, including the endoscopy suite and the Gastroenterology laboratory.

I would also like to thank Professor R D Sturrock at the Centre for Rheumatic Diseases, Glasgow Royal Infirmary, for his tremendous help in recruiting patients from the Rheumatology Outpatient Clinic, and for supervising the conduct and the progress of the studies.

The combined efforts of Dr Russell and Professor Sturrock were vital in providing the necessary funding for the studies.

In addition, I acknowledge the help of Professor F D Lee and Dr S Dahill for the histological assessments, Miss Pamela Boothman and Miss Jennifer Reid for H pylori cultures and serodiagnosis, Dr W Angerson and Mr H Beekman for their help in measuring mucosal blood flow, Dr I Nakshabendi for recruiting some patients from the Gastroenterology Clinic, Miss Jean McDonald from Medical Illustration Department,

Mrs Ruth Simpson, Mrs Carol Campbell, Mrs Christine Morran and Mrs Margaret Tosh for their secretarial assistance, Dr Stephanie Mclaughlin for the statistical analyses, Rorer Health Care and Merck, Sharpe, and Dohme for their financial support.

Finally, I would like to thank my wife Anne, children Dawod and Suzanne, parents, brothers and sisters for their encouragement and moral support.

SUMMARY OF THESIS

Summary of Thesis

This Thesis addresses several issues related to the pathogenesis of peptic ulcers in patients receiving non-steroidal anti-inflammatory drugs (NSAID).

Section I discusses the background of Thesis with respect to the mechanisms of mucosal damage by NSAID, the interaction between NSAID and *H pylori*, and the interaction between NSAID and anti-rheumatoid second-line drugs.

Suppression of mucosal prostaglandin synthesis is central to the mode of action of the vast majority of NSAID, and yet several studies have failed to find a correlation between the degree of prostaglandin inhibition and that of mucosal damage. This emphasizes the importance of other mechanisms of toxicity by NSAID such as stimulation of gastric acid secretion, interference with the mucus layer, reduction of mucosal blood flow, and the histological manifestation of chronic NSAID intake. Details of such mechanisms, in the presence or absence of *H pylori*, and their therapeutic implications are all discussed in Chapters 1 and 2. It becomes clear, as shown in Chapter 2, that NSAID and *H pylori* share several pathogenetic mechanisms which might ultimately lead to an increase in the prevalence of peptic ulcers in NSAID patients infected with *H pylori*.

In addition to taking NSAID and the possibility of being infected with H pylori, the prevalence of ulcers in rheumatoid arthritis patients might be influenced by the concurrent treatment with anti-rheumatoid second-line drugs such as gold compounds and sulphasalazine. This is described in Chapter 3, which attempts to explain the interesting observation that patients on long term gold injections appear to have a lower prevalence of both H pylori and peptic ulcers, suggesting that gold may have a toxic effect on this organism and thus protect against NSAID - induced gastric damage.

With the increasing evidence for an interaction between NSAID and H pylori, which is damaging to the gastric mucosa, the accurate diagnosis of H pylori in chronic NSAID users becomes of crucial importance. Almost all diagnostic tests for H pylori were previously evaluated in patients not taking NSAID. This subject is studied in Chapter 4, which assesses the efficacy of histology, urease enzyme activity (CLO-test), latex agglutination (Pyloriset), and an enzyme-linked immunosorbent assay, ELISA (Helico-G) in diagnosing H pylori in the presence or absence of NSAID, using culture as standard. The sensitivity and specificity of the three biopsy - related tests (culture, histology and CLO-test) were greater than 90% and not affected by NSAID intake. However, the sensitivity and specificity of Pyloriset and Helico-G were lower than those of the biopsy-related tests, and the specificity was even lower in the presence of NSAID. These results should not lead to abandoning the search for more effective serological tests because of the potential benefits of serodiagnosis, such as convenience, low cost, and early diagnosis.

Such characteristics can be very helpful especially in screening for *H pylori* and in monitoring the response to therapeutic regimes. Taking such benefits into consideration, and having accepted the validity of culture, histology and CLO-test in diagnosing *H pylori* in NSAID patients, Chapter 5 moves on to evaluate the efficacy of Biolab Malakit and Bio-Rad GAP tests, in comparison with Pyloriset and Helico-G, taking all the biopsy - related tests as standard. Bio-Rad GAP test had the highest sensitivity but the lowest specificity. The sensitivity and specificity of the other three serological tests were low, and the specificity of all the serological tests was also lower in patients taking NSAID.

The performance of the serological tests studied in Chapters 4 and 5 would limit their value in predicting peptic ulcers in patients treated with NSAID, unlike the situation in dyspeptic patients not on NSAID. The Health Assessment Questionnaire (HAQ) is an established method of assessing physical disability in patients with chronic arthritis. The prevalence of peptic ulcers might be higher in patients with debilitating arthritis because, it could be argued, their advanced disease requires more aggressive use of NSAID. This hypothesis was tested in Chapter 6, which used HAQ to classify chronic arthritic patients according to their physical disability. Patients with high HAQ scores (greater than 2.0, severely disabled by arthritis) had the highest prevalence of ulcers despite taking comparable types and doses of NSAID. The explanation for this is not fully clear, but the results might help in the process of selecting patients for prophylactic therapy against NSAID - related peptic ulcers.

Active chronic inflammation of the upper gastrointestinal tract has, for a long time, been thought to play an important role in the pathogenesis of peptic ulceration. The situation is not clear in chronic NSAID users, who are known to have a higher risk of developing ulcers. This issue is addressed in Section IV, which aims at identifying any specific histological features in the stomach, duodenum, or oesophagus of patients receiving NSAID.

Chapter 7 shows that the commonest histological abnormality in the gastric mucosa, is that of chronic superficial gastritis, followed by chemical gastritis. The latter is mostly found in chronic NSAID users who do not have a previous history of gastro-duodenal surgery. Its importance stems from its close association with NSAID related gastric ulcers.

In the duodenum, Chapter 8, a strong association is shown to exist between active chronic duodenitis, gastric metaplasia, H pylori - positive gastritis, and duodenal ulceration, regardless of NSAID intake. These results highlight the potential benefits of eradicating H pylori in the management of NSAID - related duodenal ulceration.

There are very few studies on the possible effects of NSAID on the oesophageal mucosa. Individual case reports and experimental animal studies have given conflicting results. The endoscopic part of Chapter 9 shows that oesophagitis, with or without ulcers and erosions, is commoner in NSAID patients with upper abdominal complaints than in their symptomatic counterparts not taking NSAID.

On the other hand, the histological assessments suggest that papillary elongation and basal cell hyperplasia, the major features of histological oesophagitis, are less common in the presence of NSAID. This implies that the NSAID - related oesophageal endoscopic abnormalities are more likely to reflect a localised form of damage which is comparable to other cases of pill-induced oesophagitis. The significance of such observation is that the adverse effects of NSAID on the oesophageal mucosa could be minimized by taking simple measures to prevent the retention of these tablets within the oesophagus, such as swallowing them with food and liquids.

Several studies, mostly conducted in experimental animals, have suggested that NSAID - related damage is mediated, at least in part, by mucosal ischaemia. The situation is not clear in chronic NSAID users, who have other factors that might affect mucosal blood flow, such as old age, smoking, and H pylori. The influence of these factors is studied in Chapter 10. Duodenal mucosal blood flow, measured by laser Doppler flowmetry, is lowest in chronic NSAID users who smoke, and in those with H pylori infection or duodenal ulceration. The combination of smoking and H pylori has an independent effect in lowering duodenal mucosal blood flow. Gastric blood flow does not seem to be affected by any of the above factors, However, gastric blood flow is markedly lower than that of the duodenum, and this might make it difficult to observe significant differences in gastric blood flow in response to smoking, H pylori, or gastric ulceration. In addition, the finding of lower blood flow in the stomach than the duodenum, like the relative rarity of active duodenitis and gastric metaplasia observed in Chapter 8, might

explain why gastric ulcers occur more commonly than duodenal ulcers in chronic NSAID users.

As discussed in Chapter 3, a significant number of patients with rheumatoid arthritis, receiving NSAID, also require rheumatoid disease modifying agents, known as second-line drugs. The potential benefits of such agents are frequently limited by the development of gastro-intestinal side effects, which in turn leads to the interruption or even the termination of second-line therapy. These side effects have, until recently, been studied on basis of patients' complaints only, and these are known to correlate poorly with gastro-duodenal damage in the presence of NSAID. Using endoscopy, Chapter 11 investigates the prevalence of peptic ulcers in rheumatoid arthritis patients taking NSAID with or without second-line drugs. Patients receiving NSAID only, or together with sulphasalazine, penicillamine or hydroxychloroquine, appear to have a similar prevalence of peptic ulcers. However, the chronic intake of gold injections plus NSAID is associated with a smaller number of ulcers than that of NSAID only. This finding is likely to influence the choice of second-line agents in rheumatoid arthritis. Also, the absence of a rise in the prevalence of ulcers in the sulphasalazine group is reassuring: upper abdominal symptoms, frequently seen in such patients, should not necessarily lead to stopping sulphasalazine therapy. Indeed, these symptoms disappear in the majority of cases with minor adjustment of the dose of sulphasalazine, and with the temporary use of anti-emetics early in the course of sulphasalazine therapy.

Chapter 12 attempts to explain some of the endoscopic findings of Chapter 11, with respect to the apparently favourable effects of gold on the gastric mucosa. Compared with patients treated with NSAID only or with sulphasalazine, the prevalence of both peptic ulcers and H pylori is lower in patients receiving long term gold injections. This confirms the findings of Chapter 11, and suggests that the apparently protective effect of gold against NSAID - related ulcers is probably mediated, at least in part, by the effect of gold on gastric H pylori.

In conclusion, this Thesis consists of a group of studies aimed at clarifying several aspects of the pathogenesis of NSAID - related peptic ulcers, and it is hoped that the findings will assist in the management of such lesions in chronic NSAID users.

SECTION 1
BACKGROUND OF THESIS

INTRODUCTION

NSAID are known to inhibit mucosal prostaglandin synthesis, stimulate gastric acid secretion, interfere with mucosal blood flow and the mucus layer, and increase the mucosal permeability. These activities are reviewed in Chapter 1, which also considers the therapeutic approaches to NSAID - related damage. The recent entry of H pylori into the debate of peptic ulcer aetiology makes it essential to consider the possible role of this organism in the pathogenesis of ulcers in patients taking NSAID. Chapter 2 reviews the pathogenetic mechanisms common to both NSAID and H pylori. The impact of the concurrent intake of anti-rheumatoid second-line drugs on both H pylori and NSAID - related ulcers is also discussed in Chapter 3.

CHAPTER 1

1. MECHANISMS OF NSAID - RELATED PEPTIC DAMAGE AND THEIR IMPLICATIONS FOR MANAGEMENT.

1.1 INTRODUCTION

Despite the wide acceptance of the strong association between non-steroidal anti-inflammatory drugs (NSAID) and peptic ulcer disease, there are several issues related to the pathogenesis and management of this disease that are at risk of being over-looked or accepted unchallenged. This chapter discusses such issues and considers their implications for the management of NSAID related peptic ulceration.

1.2 INTERFERENCE WITH PROSTAGLANDIN SYNTHESIS BY NSAID

Inhibition of prostaglandin (PG) biosynthesis is the most famous activity of NSAID, and it is thought to account for their anti-inflammatory and analgesic properties. It was, and still remains, very desirable if not vital in the treatment of arthritic disorders. It has subsequently been considered a significant disadvantage after associations have been made between NSAID - related peptic damage and gastro-duodenal mucosal PG inhibition. Before these points are discussed in further details, there are certain features related to the effect of NSAID on PG production that should be noted.

NSAID, such as indomethacin, suppress PG biosynthesis by inhibiting cyclo-oxygenase activity, the enzyme which is responsible for converting arachidonic acid substrate to PG intermediates. NSAID are considered not to have any major effects on the enzymes involved in the lipoxygenase system, and therefore the production of the leukotrienes would only be affected by these drugs to the extent that further arachidonic acid substrate would be available for leukotriene biosynthesis on account of the decreased PG production.

NSAID appear to vary in their potencies to inhibit PG synthesis (1). Indomethacin and naproxen are powerful inhibitors, phenylbutazone is moderate, while salicylic acid, aspirin, is only a weak inhibitor. Other drugs act as pro drugs that show activity through their metabolites: one of these agents is sulindac whose sulphide metabolite is able to exert a potent inhibiting effect on PG synthesis (2).

It is also interesting to note that NSAID may show selectivity in terms of which PGs are mainly affected. Previous observations with sheep vesicular tissue incubated in Cu^{+2} containing medium indicated that salicylic acid caused a preferential reduction in PGE_2 synthesis relative to that of PGF_2 (3). In contrast, indomethacin suppressed the production of PGE_2 , PGE_1 , and PGF_2 to the same extent. More recently, it has been shown that etodolac, a derivative of pyranocarboxylic acid, might selectively spare gastric and duodenal mucosal PGs despite its capacity to suppress synovial PG synthesis (4,5).

While it is generally agreed that NSAID suppress gastric mucosal

PGs, knowledge in this field has largely relied on studies conducted in animals or healthy humans given short courses of aspirin or indomethacin (1-3, 6-8). As shown above, what applies to these two agents, aspirin and indomethacin, might not necessarily apply to the rest of NSAID, and species-specific responses should not be ignored. Also, very little is known about the behaviour of duodenal mucosal PGs in patients treated with NSAID as the vast majority of studies have investigated gastric PGs only. More importantly, no correlation has been found between the degree of PG inhibition and that of NSAID-induced peptic damage (5,8). These reservations do not necessarily minimize the acknowledged importance of PGs in maintaining the integrity of the gastro-duodenal mucosa (9): they rather emphasize the importance of considering other mechanisms of injury by NSAID in addition to their effect on PG production, as discussed below.

1.3 OTHER MECHANISMS OF MUCOSAL TOXICITY BY NSAID

Aspirin (10), indomethacin (11), and fenoprofen (12) were found to cause disruption of the gastric mucosal barrier in a manner that allows back diffusion of hydrogen ion with its damaging consequences. Aspirin and indomethacin also increase basal (13) and maximally stimulated gastric acid secretion (14,15) which may contribute to their unwanted effects on the gastric and duodenal mucosa.

Several studies have suggested that NSAID can affect the rate of secretion and/or the characteristics of the mucus layer in the stomach. Aspirin (16) and indomethacin (17) were found to inhibit

mucus secretion. It was also suggested that aspirin could increase pepsin-mediated proteolysis of mucus, decrease mucus viscosity, and increase the permeability of mucus to hydrogen ion (18). Indomethacin was also found to inhibit active bicarbonate secretion by the gastric mucosa (19). In addition, indomethacin was found to decrease gastric mucosal blood flow (14).

Another mechanism that received insufficient attention in the past relates to the possibility that bacteria could mediate NSAID toxicity. In the intestine it was observed that the ulcerogenic effect of NSAID could be reduced by antibiotic treatment (20). Germ-free animals were also found to be resistant to indomethacin-induced intestinal lesions (21). It was postulated that enteric bacterial β -glucuronidase hydrolyses the acylglucuronides of NSAID released into the intestinal tract from bile, and the free acids then irritate the mucosal surface (20). Interest in these important observations has been revived by the recent entry into the argument of another organism: *Helicobacter (H) pylori*. It is not clear whether NSAID interact with *H pylori* in the gastric mucosa. However, studies conducted by the author have shown that human gastric antral mucosal specimens exposed to a mixture of indomethacin plus *H pylori* culture filtrate have lower PGE_2 values and histological viability grades than biopsies incubated with indomethacin or the culture filtrate alone (22). More recently, it has been found that the prevalence of NSAID-related ulcers is higher in patients with chemical gastritis or *H pylori*-related gastritis (23, Chapter 7). An interaction may, therefore, exist between NSAID and *H pylori*, and this seems to be damaging to the gastric mucosa. The interaction between NSAID and *H pylori* is discussed in more detail in Chapter 2

of this Thesis.

It is worth noting that a degree of overlapping might exist between two or more of the above mechanisms of NSAID toxicity. For example, the effect of indomethacin on mucosal blood flow might not be totally independent of that on PG formation. However, despite the capacity of PGs to inhibit gastric acid (24,25), the effects of NSAID on acid secretion is not necessarily mediated through their suppression of mucosal PGs: recent studies have suggested that NSAID bypass the H_2 and muscarinic receptors and interact with secretagogues at a locus between the catalytic subunit of adenylate cyclase activation and the proton pump (26). The potentiation of secretagogue-stimulated acid secretion by non-salicylate NSAID has been found to be dependent on calcium (26).

It is clear, therefore, that in addition to their effects on PG synthesis, NSAID have several other activities which have to be addressed when management of NSAID-related damage is considered.

1.4 IMPLICATIONS FOR MANAGEMENT

The above discussion would suggest that there might be justification, which is at least theoretical, for the use of any class of anti ulcer drugs in the management of ulcers induced by NSAID.

Antacids that contain aluminium hydroxide or aluminium phosphate have been shown to protect the rat's gastric mucosa against necrotizing conditions (27,28). Aluminium hydroxide was found to

increase PGE₂ release by the gastric mucosa in rats (29). A local mechanism must be responsible for these activities of aluminium hydroxide since it is poorly absorbed. Also, protection through other mechanisms cannot be entirely excluded: these might include adsorption of bile acids, inactivation of pepsin or stimulation of mucus production (29). The value of antacids in treating or preventing NSAID-related damage in man has not been established yet.

Carbenoxolone sodium was previously reported to increase the formation of gastric mucosal PGE₂ (30) and to inhibit thromboxane B₂, the stable metabolite of thromboxane A₂ (31). Despite these highly desirable properties in the context of treating ulcers caused by NSAID, carbenoxolone is not widely used in clinical practice because of its cardiovascular side effects.

Tripotassium-dicitrate bismuthate (colloidal bismuth) was shown to stimulate mucosal PGI₂-like activity and to protect the rat's gastric mucosa against 85% ethanol, 0.2N sodium hydroxide, or acidified indomethacin (32). Colloidal bismuth subcitrate was also found to reduce aspirin-induced microbleeding in man despite marked suppression of gastric mucosal PGs by aspirin (33). Recently, interest has been renewed in colloidal bismuth in light of its effect on H pylori, and might increase further because of the apparent interaction between NSAID and H pylori (22,23).

Despite the conflicting reports on the capacity of sucralfate to stimulate mucosal PGE₂ release (34,35), it is generally accepted that this complex molecule behaves as a mild irritant by forming a

gelatinous mucoepithelium coat consisting of necrotic epithelial cells and mucus which in turn acts as a shield against acid and pepsin (36). The application of these observations to NSAID-related damage in man remains to be confirmed.

In recent years, several important clinical trials have been reported on the value of H₂-antagonists and prostaglandin analogues in the treatment or prevention of peptic ulcers in patients receiving NSAID. Conflicting results were reported with cimetidine (37,38). Rantidine was found to prevent NSAID-related duodenal but not gastric ulceration (39). More recently, ranitidine was shown to heal 63% of gastric and 84% of duodenal ulcers after 8 weeks of treatment, compared with 79% and 92% at 12 weeks, respectively, in patients who continued to take NSAID (40).

In a double-blind, placebo-controlled trial Graham et al have shown that misoprostol, PGE₁ analogue, significantly reduced the frequency of gastric ulcer development in patients treated with NSAID (41). The importance of these findings and those of Ehsanullah et al would justify a comparison between the two studies (39,41): In addition to the differences outlined in Table 1, it is not clear why NSAID patients in the Ehsanullah's study (39) developed almost equal numbers of ulcers in the stomach and duodenum given the fact that gastric ulcers tend to be commoner than duodenal ulcers in patients receiving NSAID (42). Since the two studies (39,41) were different in their design and in the demographic details of their patients, it is difficult to perform a direct comparison between ranitidine and misoprostol on basis of their findings. This makes it essential for the two agents to be compared in one single

trial. When such comparison was made between misoprostol and cimetidine, both agents were highly protective in the duodenum but misoprostol was more effective in protecting against tolmetin-induced gastric injury in healthy volunteers (43). It might be tempting to consider giving the individual patient treated with NSAID a combination of an H₂ antagonist, to relieve dyspeptic symptoms and prevent duodenal ulcers, and misoprostol to prevent gastric ulceration. As a matter of fact, the author has seen patients who have already been treated with such combination. To do so is difficult to justify not only on basis of cost but also because misoprostol has been shown to protect against duodenal as well as gastric ulcers (41,43,44).

However, it is very important to realise that the value of H₂ antagonists has not been adequately tested as shown in Table 1, and their use should not therefore be ruled out, at least in treating NSAID-induced peptic ulceration (40). A direct comparison is indicated between ranitidine and misoprostol and more information is needed about the value of other agents such as famotidine and omeprazole in treating or preventing ulcers related to NSAID. Also, the impact of these treatment strategies on the complications of NSAID-related lesions remains to be investigated.

In conclusion, NSAID have multiple effects on the gastro-intestinal mucosa in addition to inhibition of mucosal PG synthesis. The understanding of such effects is essential for designing the appropriate lines of management of ulcers related to NSAID.

Table 1: Prevention of NSAID-Related Peptic Ulcers By Ranitidine (Ehsanullah et al, 39) Or Misoprostol (Graham et al, 41): Comparison Of The Design And Findings Of The Two Studies. No (%)

	<u>Ranitidine</u>	<u>Misoprostol</u>
	<u>Study</u>	<u>Study</u>
No of patients	263	420
Osteoarthritis	197 (75%)	420 (100%)
Rheumatoid arthritis	66 (25%)	-
Smokers	94 (36%)	63 (15%)
NSAID used	Naproxen	Naproxen
	Piroxicam	Piroxicam
	Diclofenac	Ibuprofen
	Indomethacin	-
Duration of NSAID treatment	8 weeks	1 year
Duration of study	8 weeks	12 weeks
Lesions studied	erosions	ulcers >3mm
	ulcers >5mm	ulcers >5mm
Prevention of ulcers:		
Gastric	No	Yes
Duodenal	Yes	Not studied
Symptomatic relief	Yes	No
Significant side effects		
of study drug	Nil	Diarrhoea (25-39%)

CHAPTER 2

2 THE INTERACTION BETWEEN NSAID AND H PYLORI

2.1 SUMMARY

Recent studies have suggested that *Helicobacter pylori* and non-steroidal anti-inflammatory drugs are capable of interfering with various protective mechanisms in the gastro-duodenal mucosa. While NSAID are recognised for their acid stimulating activity, the effect of *H pylori* on gastric acid secretion remains highly speculative despite its association with hypergastrinaemia. Both *H pylori* and NSAID have, however, been shown to influence the production rate and/or the quality of the mucus layer, gastric cyclic AMP, mucosal prostaglandins, blood flow, and platelet-activating factor. Characteristic histological abnormalities have also been identified. In addition, NSAID-related peptic ulcers seem to develop more commonly in patients infected with *H pylori* despite the apparent reduction in the prevalence of these organisms in chronic NSAID users. It remains to be seen whether the eradication of *H pylori* would reduce the frequency of NSAID induced peptic ulcers or prevent their recurrence.

2.2 INTRODUCTION

Helicobacter (H) pylori and non-steroidal anti-inflammatory drugs (NSAID) are probably the commonest known exogenous factors in the aetiology of peptic ulcer disease. This Chapter describes the pathogenetic mechanisms common to H pylori and NSAID and discusses the possibility of a synergistic relationship between them. The importance of such a relationship, if present, relates to its potential to provide a new therapeutic approach to the common, but yet unresolved, problem of ulcers induced by NSAID.

2.3 EFFECT OF NSAID AND H PYLORI ON GASTRIC ACID SECRETION:

Because of the widely accepted importance of gastric acid in the pathogenesis of ulcers in general, it was only natural for many workers to investigate the possible effects of NSAID and, more recently, H pylori on acid secretion. NSAID (indomethacin and aspirin in particular) were found to increase basal and maximally stimulated gastric acid secretion (13-15). NSAID seem to bypass the H₂ and muscarinic receptors and interact with secretagogues at a locus between the catalytic subunit of adenylate cyclase activation and the proton pump (26). The potentiation of secretagogue - stimulated acid secretion by non-salicylate NSAID has also been found to be dependent on calcium (26).

The situation is not so well defined in the case of H pylori. Because of its association with hypergastrinaemia, it was speculated

that H pylori could increase the parietal cell mass that is characteristic of duodenal ulcer patients (45). To date, the evidence for an increase in gastric acid secretion by H pylori, has however, been lacking. On the contrary, there is a consensus that acute exposure to H pylori causes hypochlorhydria (46-50).

Recent evidence indicates that H pylori associated hypergastrinaemia might not be directly related to the function of the parietal cells (51), the number of antral G cells (52) or to the bacterium's urease activity (53). It is more likely to be related to local inflammation (52-54) and/or products of the T lymphocyte such as interleukin-2 and gamma-interferon (55). Gastric acid secretion in chronic NSAID users who are infected with H pylori has not been studied, but indomethacin has been shown to potentiate the inhibitory effect of H pylori protein on gastric fundic cyclic AMP, which in turn mediates acid secretion, in vitro (56). This might explain, at least in part, the tendency of many arthritics to develop a degree of hypochlorhydria (57), and the limitations of acid inhibition in the management of NSAID related ulcers (39,43).

2.4 THE MUCUS LAYER

The ability to change the characteristics of the gastric mucus layer is common to both NSAID and H pylori. Aspirin (16) and indomethacin (17) were found to inhibit mucus secretion. Aspirin can also increase pepsin-mediated proteolysis of mucus, decrease mucus viscosity, and increase the permeability of mucus to hydrogen ion (18). Indomethacin was shown to inhibit active bicarbonate secretion by the gastric mucosa (19). It was also suggested that NSAID could cause disruption of the gastric mucosal barrier which in

turn allows back diffusion of hydrogen ion with its damaging results (10-12).

Similarly, it has recently been shown that incubation of H pylori culture filtrate with gastric mucus could lead to a gradual loss of mucus viscosity (58), which might impair its ability to retard the diffusion of hydrogen ions. It was concluded that the degenerative changes produced in the gastric mucus gel by H pylori might be a contributing factor in the pathogenesis of gastritis and peptic ulcers (58)

2.5 MUCOSAL PROSTAGLANDINS:

It is generally accepted that NSAID, with few exceptions (5), are capable of suppressing gastric and duodenal mucosal prostaglandin (PG) synthesis, although no correlation has been found between the degree of PG inhibition and the endoscopic abnormalities (5,8).

Because of its strong association with gastritis and neutrophilic infiltration, H pylori is expected, at least in theory, to stimulate PG production, as human neutrophils and macrophages are capable of synthesising PGs (59). Also, gastritis like any other inflammation irrespective of H pylori status is associated with elevated PG values (60,61). It was surprising, therefore, to find that patients with H pylori related gastritis had PG values similar to those without H pylori (61-63). This led to speculation that H pylori infection might result in at least a partial block in PG synthesis at the level of the neutrophils, mucosal cells, or both (61). Indeed, some workers

reported reduced levels of PGI₂ stable metabolites in patients infected with H pylori (64), but this was not confirmed by others (61-63). It was also interesting to find that the combination of indomethacin and H pylori culture filtrate reduced gastric antral mucosal PGE₂ and epithelial viability to a greater degree than indomethacin alone, in vitro (22). This in turn might suggest a synergistic relationship between H pylori and NSAID in causing mucosal damage.

2.6 MUCOSAL BLOOD FLOW:

Several studies have demonstrated that short courses of NSAID could reduce gastric mucosal blood flow in the laboratory animals (14,65,66). The situation is not clear in patients on long term NSAID, who could have other factors that might alter blood flow. Also, very little is known about the duodenal microcirculation in such patients. However, it has been shown that duodenal, but not gastric, mucosal blood flow is lowest in NSAID patients who smoke, or those with duodenal ulcers or H pylori (67, Chapter 10); no correlation could be found with age or sex. The data in Chapter 10 also show that the suppressive effect of smoking and H pylori on duodenal blood flow, in NSAID patients, was independent of ulceration and other demographic variables. Although such findings do not dispute the capacity of NSAID to lower mucosal blood flow in general, they might represent another aspect of the synergistic relationship thought to exist between NSAID and H pylori, at least with respect to duodenal blood flow.

2.7 HISTOLOGICAL AND ENDOSCOPIC ABNORMALITIES:

Active chronic gastritis is the classical histological picture found in association with *H pylori* in the majority of infected subjects. It is best remembered as the main abnormality seen after the deliberate ingestion of the organisms in order to fulfil Koch's postulates for the pathogenicity of *H pylori* (46,47), and in epidemic gastritis (48). It is also the commonest histological finding in patients treated with NSAID (23, Chapter 7). Neutrophilic infiltration, common to most cases of NSAID and *H pylori* gastritis, could be related to the capacity of NSAID and *H pylori* to stimulate the production of the platelet-activating factor (PAF) (68,69). In addition to its ulcerogenic actions (68), PAF might be involved in the aggregation and activation of neutrophils (70), which might subsequently contribute to ulcer formation: neutropenic rats are less likely to develop gastric mucosal injury by indomethacin or naproxen (70). However, these interesting observations might not apply to patients with chemical gastritis, another histological abnormality characteristic of chronic NSAID intake (23,71 Chapter 7). Not unlike bile reflux gastritis in patients with a history of gastroduodenal surgery (72), it is characterised by foveolar hyperplasia, vasodilatation, oedema, lack of inflammatory cells, and the presence of muscle fibers in the lamina propria (23,71-74, Chapter 7). *H pylori* is very rare in cases of chemical gastritis (23, 71-74, Chapter 7), and the histological picture can be found in about 25% of chronic NSAID users (23,71 Chapter 7). The importance of such entity stems from the positive correlation between the histological chemical scores and the degree of endoscopic damage: NSAID related peptic ulcers, measuring 5mm in diameter, were found more commonly in patients

with chemical gastritis or H pylori positive gastritis than in those without either of these conditions (23, Chapter 7). This is in agreement with another study (75) which found a greater number of gastric ulcers in NSAID patients infected with H pylori compared with patients either taking NSAID or infected with H pylori. The prevalence of NSAID related ulcers in H pylori positive patients has also been found to be twice that in H pylori negative individuals, but the differences were not significant due to the small numbers of patients and ulcers studied (76). The same study (76) found a greater number of submucosal haemorrhages and erosions in H pylori negative NSAID users, unlike the results of another report which suggested that the presence of H pylori had no influence on the prevalence of such lesions following the acute administration of naproxen or aspirin in healthy volunteers (77).

The interpretation of the findings related to submucosal haemorrhages or erosions is made difficult by the chronic nature of H pylori infection, its association with ulcers, and the lack of evidence that such minor lesions can really progress to ulcers. Another possible explanation for the differences in the findings of the above studies (23,75-77, Chapter 7) is the failure to correct for the presence of chemical gastritis, which appears to act as an independent factor, and the use of serological tests of unproven value in patients treated with NSAID. Diagnostic titres of H pylori IgG antibodies could still be detected in patients with chemical gastritis, with its high prevalence of NSAID ulcers, despite the failure to confirm the presence of the organisms by culture, histology, or urease activity (78, Chapter 4). Also, serology might not be reliable enough in predicting the presence of H pylori or

ulcers in NSAID patients, unlike other categories of patients (79,80), because of the following: Firstly, NSAID ulcers can be completely asymptomatic (81,82, Chapter 11); Secondly, the specificity of some IgG tests might be low in patients receiving NSAID (78, Chapter 4); Thirdly, despite the apparent synergistic relationship between H pylori and NSAID, the intake of such drugs might be associated with a reduced prevalence of H pylori infection (23,78,83-85, Chapters 4,7): this could be due to a direct toxic action, or indirectly related to the increase in acid secretion (13-15,26) and the interference with the mucus layer (10-12,16-19) rendering the natural habitat of H pylori organisms unsuitable for their survival.

The virulence of H pylori strains might also explain some of the differences in the reported rates of ulcer formation. Western blotting analysis of systemic IgG or IgA responses to H pylori has shown the antigenicity of 110-120 kDa, 89 kDa, 61 kDa, 54 kDa and 31 kDa proteins, although there is substantial variability amongst subjects (86,90). The 120 kDa protein, which is recognized systemically in 83% of H pylori positive subjects, is a surface protein not expressed in some H pylori strains (88).

It has been suggested that 120 kDa positive strains have pathogenetic features associated with active gastritis and peptic ulceration (90). Infection with 120 kDa negative strains might explain why peptic ulceration develops in only a proportion of subjects infected with H pylori (90). A vaculating cytopathic agent has also been described in some strains of H pylori (91). It is not,

however, known whether these important findings would apply to patients receiving long term NSAID.

2.8 THERAPEUTIC CONSIDERATIONS:

The role of eradicating H pylori in the healing or the prevention of NSAID related peptic ulcers has not been studied. However, in addition to their antibacterial activity, agents currently used in the treatment of H pylori infection were previously found to possess properties relevant to the mechanisms of NSAID induced mucosal damage. Colloidal bismuth subcitrate was shown to stimulate gastric and duodenal alkaline secretion through a prostaglandin dependent mechanism (92). The same agent was also found to have a protective action against aspirin-induced microbleeding, and this protection occurred despite a marked suppression of mucosal prostaglandin production (33). Also, it was demonstrated that the ulcerogenic effect of NSAID could be reduced in animals and humans by antibiotic treatment (20,93), and that germ-free animals were resistant to indomethacin-induced intestinal lesions (21).

The common nature of NSAID related ulcers in association with H pylori colonization, especially in the elderly, should act as an incentive to devise an effective therapeutic approach. In 1983, enough NSAID were made available to treat almost 3 million people daily in the USA (94), and 22 million prescriptions were issued for such agents in the UK (95). About half of the NSAID prescribed were for the over 60's (95). H pylori infection can be identified in about 80% of people of this age group (96) although in the presence of NSAID the prevalence of H pylori might fall to 30-50%, according

to data collected by the author between 1987-1992 (23,67,78)

It is estimated, therefore, that between 25-40% of all NSAID users and 30-50% of elderly patients taking NSAID could be infected with H pylori, and as a result, are at special risk of developing peptic ulcers. The management of ulcers in this large number of patients is not only essential but might also need to be different from that of other types of ulcers. It's effect on ulcer complications, especially in the elderly, also needs to be clarified.

Despite the evidence for a low prevalence of H pylori in chronic NSAID users, it has to be emphasised that the possible use of NSAID to eradicate H pylori should not be considered, knowing that such eradication might involve the impairment of the mucus layer with its damaging consequences.

In conclusion, the evidence for a synergistic relationship between NSAID and H pylori, despite being contested should not be ignored because of its potential therapeutic implications. Also, the suggestion that NSAID might influence the prevalence of H pylori would indicate that the two factors are uncomfortable partners in peptic ulcer disease.

CHAPTER 3

3. NSAID-RELATED ULCERS AND ANTI-RHEUMATOID SECOND-LINE DRUGS

3.1 INTRODUCTION

Many patients with rheumatoid arthritis need both NSAID and second-line drugs for the treatment of their arthritis. Gastro-intestinal side effects, previously studied on basis of patients symptoms only, are the commonest cause of interrupting or even discontinuing second-line therapy. The issue is also complicated by the concurrent intake of NSAID. The studies included in Section VI suggest that patients receiving NSAID and intramuscular gold injections have a lower prevalence of peptic ulcers than those treated with NSAID only or with other second-line drugs including sulphasalazine, penicillamine, and hydroxychloroquine. The prevalence of *H pylori* was also lower in patients treated with gold. In light of these findings, Chapter 3 reviews the mechanisms of interaction between gold and the gastric mucosa, and discusses their implications for the management of NSAID-related ulcers in patients with rheumatoid arthritis.

3.2 HISTORICAL BACKGROUND

As some rheumatologists are rethinking the value of gold therapy in rheumatoid arthritis (RA) (97-100) an increased interest in this precious metal has occurred amongst gastroenterologists (82,101-105, Chapter 11) because of its potential benefits in peptic ulcer disease.

The varying degrees of interest in gold therapy, noted over the years, has each time been linked to its antibacterial activity. Nearly a century after gold was used in the treatment of tuberculosis (106) and 60 years after its first use in the therapy of rheumatoid arthritis (107), then thought to be tuberculous in origin, the drug is now being reconsidered in the treatment of another infection: *Helicobacter (H) pylori* (101-105), which has been linked to peptic ulcer disease.

3.3 MECHANISMS OF INTERACTION BETWEEN GOLD AND THE GASTRIC MUCOSA, AND THEIR THERAPEUTIC IMPLICATIONS

Not unlike its anti-rheumatoid activity, the effect of gold on the gastric mucosa is still poorly understood. Its apparent protective effects on the stomach are in contrast to those on the colonic mucosa where it may cause colitis (108). Gold may affect factors involved in the pathogenesis of peptic ulcer disease including gastric acid, *H pylori*, and NSAID; and the protective factors, including mucosal prostaglandins, bicarbonate secretion, and the mucus layer. These factors cannot always be considered separately, as the individual

patient might be infected with H pylori, receiving an NSAID with decreased gastric prostaglandin synthesis, all of which increase the risk of ulcer formation.

The effect of gold on gastric acid secretion has not been studied. Despite its importance, the role of acid in the pathogenesis of peptic ulcers in rheumatoid patients might not be critical, as many RA patients tend to have hypochlorhydria (57). Also, the effect of gold on gastric prostaglandins, mucus, and bicarbonate secretion remains speculative. Since gold and bismuth are classified close to one another in the periodic table of elements, it might be justifiable to suppose that their mode of action might be similar (82,101, Chapter 11). Colloidal bismuth subcitrate was found to have a protective effect against aspirin-induced gastric microbleeding, and this protection occurred despite suppression of mucosal prostaglandin production by aspirin (33). In addition, colloidal bismuth subcitrate was shown to stimulate gastric and duodenal alkaline secretion through a prostaglandin dependent mechanism (92). Whether gold possesses similar favourable properties to bismuth needs to be investigated.

More is known about the interaction between gold, H pylori, and NSAID than that between gold and other factors such as acid, prostaglandins, and bicarbonate secretion. The low prevalence of H pylori in rheumatoid patients treated with gold compounds might be due to either a direct bactericidal effect, as shown by in vitro studies (101), or inhibition of H pylori urease activity (102), or both. The failure of some studies to demonstrate any significant effect of gold on H pylori colonization (103) could be explained by

the difference in the methodology of detecting H pylori organisms: unlike culture, histology, and the CLO-test, which involve gastric biopsies, the specificity of some serological tests for H pylori might be less reliable in RA (78, Chapter 4). The immunomodulatory activity of gold is another mechanism by which this agent might influence the survival of H pylori, and this is of particular relevance to patients with RA. It has been observed that immuno-deficient patients are less likely to be infected with H pylori possibly due to impaired host cellular immune responses (109,110). Sulphaslazine, known to have anti-arthritic activity comparable to that of gold, does not seem to affect the prevalence of H pylori (105, Chapter 12).

Chronic suppression, rather than eradication of H pylori by gold compounds, could also explain the small number of cases of detectable organisms in biopsies taken from chronic gold users (105). This implies that H pylori might recur or become more easily detectable upon the withdrawal of gold therapy. The presumed effect of intramuscular gold on H pylori (105) is thought to be due to the distribution of gold into the gastric mucosa, gastric secretions, or both. There have been no studies on gastric tissue or juice levels of gold, but it could be speculated that such levels might be higher if oral gold preparations are used, which in turn might be more effective against H pylori.

It has to be emphasized that the use of current gold preparations to eradicate or suppress H pylori in non-arthritic patients cannot be justified, at this stage. This is because of their relative expense, potential toxicity, and the availability of safer anti-H pylori drugs such as bismuth compounds, antibiotics, and

more recently, the proton-pump inhibitors.

In conclusion, gold continues to be an intriguing precious metal which has fascinating and complicated activities in biological systems. An understanding of its effects on *H pylori* may yield useful information germane to its mode of action in RA. Its gastroprotective properties are a surprising and a welcome bonus to RA patients whose stomachs are otherwise exposed to the insults of their antirheumatic therapy.

SECTION II
AIMS OF THESIS

SECTION II

AIMS OF THESIS

It is obvious, as discussed in Chapters 1-3, that the pathogenesis of NSAID-related peptic damage is still not fully understood. There are several areas that need further clarification; these include the possible interaction between NSAID and H pylori, the effect of NSAID on the detection of H pylori, the histological features of NSAID-related damage, the effect of NSAID on mucosal blood flow, and the possible interaction between NSAID and anti-rheumatoid second-line drugs.

The suggestion that NSAID-related ulcers occur more commonly in patients infected with H pylori (Section I), makes it essential to be able to detect these organisms accurately in patients taking NSAID. The aim of Chapter 4 is, therefore, to assess the efficacy of culture, histology, CLO-test, Helico-G, and Pyloriset tests in identifying H pylori infection, in the presence or absence of NSAID, using culture as standard.

Given the potential advantages of diagnosing H pylori by serology, including speed, convenience, low cost, and labour saving, Chapter 5 aims at evaluating a total of four serological tests in patients taking NSAID. These include Biolab Malakit, Bio-Rad GAP IgG, Helico-G, and Pyloriset tests. All three biopsy-related tests are used as standard.

Chapter 6 assesses the ability of the Health Assessment Questionnaire to predict NSAID-related ulcers in patients with

arthritis. This is based on the hypothesis that patients with debilitating joint disease might be at a greater risk of developing NSAID-related ulcers.

Active chronic inflammation of the upper gastro-intestinal tract has always been thought to play an important role in the pathogenesis of peptic ulcer disease. The situation is not clear in chronic NSAID users. Section IV aims at identifying the histological abnormalities associated with NSAID intake in the stomach (Chapter 7), duodenum (Chapter 8), and the oesophagus (Chapter 9).

The interference with gastric mucosal blood flow is another important side-effect of NSAID. Little is known about the duodenal microcirculation in NSAID patients and it is not clear whether the mucosal blood flow is affected by factors such as age, smoking, H pylori, and active ulceration. Chapter 10 aims at investigating the influence of these factors on gastric and duodenal blood flow in patients receiving long term NSAID.

Since many patients with rheumatoid arthritis are treated with both NSAID and second-line drugs, it might not be easy to explain the source of gastro-intestinal complaints in such patients. The aim of Chapter 11 is to identify the endoscopic abnormalities in patients receiving NSAID only, or with gold, sulphasalazine, penicillamine, and hydroxychloroquine. In addition, Chapter 12 investigates the prevalence of both ulcers and H pylori in patients treated with NSAID only or with gold and sulphasalazine: the latter two agents

are known to have comparable anti-rheumatoid effect, and were originally used for their presumed anti--microbial activity, in the days when rheumatoid arthritis was thought to be of bacterial origin.

SECTION III
DIAGNOSTIC STUDIES

INTRODUCTION

This section investigates the efficacy of a total of seven tests used in the diagnosis of H pylori in the presence or absence of NSAID: 3 biopsy-related tests (culture, histology, CLO-test), and 4 serological tests (Pyloriset, Helico-G, Biolab Malakit, and Bio-Rad GAP IgG). It also tests the capacity of the Health Assessment Questionnaire to predict NSAID-related ulcers in arthritic patients taking NSAID.

CHAPTER 4

4 INFLUENCE OF NSAID ON THE EFFICACY OF DIAGNOSTIC TESTS FOR H PYLORI

4.1 SUMMARY

Background: The efficacy of various tests used in the diagnosis of *Helicobacter pylori* has not been fully established. Also, it is not known whether the yield of such tests can be influenced by non-steroidal anti-inflammatory drugs.

Aims: To evaluate the efficacy of culture, histology, CLO-test, Helico-G and Pyloriset tests in diagnosing *H pylori* in the presence or absence of NSAID.

Patients and Methods: Of 134 patients studied, 75 took NSAID and 59 did not. At endoscopy, biopsies were taken for culture, histology and CLO-test. Blood was also taken for ELISA (Helico-G) and latex agglutination (Pyloriset) tests.

Results: The sensitivity, specificity, and predictive values of histology and CLO-test, compared with culture, ranged between 90-97% regardless of NSAID intake. In patients not taking NSAID, Helico-G had a sensitivity of 75% ($p < 0.05$) and a specificity of 61%; Pyloriset's sensitivity and specificity were, respectively, 63% ($p < 0.05$) and 67%. In patients taking NSAID the sensitivity of Helico-G was 81% and its specificity was 45% ($p < 0.05$); Pyloriset had

a sensitivity of 61% ($p < 0.05$) and specificity of 50% ($p < 0.05$).

Conclusion: These findings suggest that *H pylori* is more reliably diagnosed by culture, histology and CLO-test than by the serological tests used in this study, especially in patients treated with NSAID.

4.2 INTRODUCTION

The increasing realization of the importance of *Helicobacter* (*H*) *pylori* in the development, treatment, and recurrence of peptic ulceration (111-115) has made the identification of these organisms an essential part of the investigation and management of peptic ulcer disease. Several tests have been proposed including culture, histology, urease enzyme activity, and serological tests (79, 116-121). The value of such methods in general and the serological tests in particular has not been fully established. Also, their application to patients treated with non-steroidal anti-inflammatory drugs (NSAID), who are at a particular risk of developing peptic damage, has not been adequately investigated.

The aim of this study was to evaluate the efficacy of culture, histology, urease activity (CLO-test), enzyme linked immunosorbent assay (ELISA, Helico-G), latex agglutination test (Pyloriset) in diagnosing *H pylori* in the presence or absence of NSAID

4.3 PATIENTS AND METHODS

Patients, over the age of 18 years, were recruited from the

Gastroenterology and Rheumatology clinics. NSAID had to be taken for a minimum of 4 weeks prior to endoscopy. Patients were excluded if they had a history of gastric surgery or if they had received antibiotics or ulcer healing drugs within 2 weeks of endoscopy.

Informed consent was obtained and endoscopy carried out, using 3-6mg midazolam intravenously for sedation, within 3 hours of taking 10ml of venous blood for serology tests. Biopsies were taken from healthy looking mucosa in the gastric antrum and at least 2cm away from the ulcer edge, in patients with ulcers, for culture (1 biopsy), histology (2 biopsies), and CLO-test (1 biopsy). In patients with ulcers, biopsies were also taken from the ulcer edge and base to exclude malignancy which, if found, excluded patients from the final analysis.

H pylori was detected by culture and histology as previously described (61). Gastritis was classified according to the Whitehead system modified to cover chemical and lymphocytic gastritis (23,72,122,123, Chapter 7).

Urease activity was tested for by inserting one antral biopsy, immediately after it was obtained, into the gel pellet of the CLO-test slide (Delta-West Ltd, Bentley, Western Australia). The slides were reviewed for the positive red colouration at 3 and 24 hours.

Serum IgG H pylori antibodies were measured using a quantitative commercial IgG ELISA (Helico-G) according to the manufacturer's instructions (Porton Cambridge, Maidenhead, UK). In summary, a

standard curve was prepared and 100ul serum aliquotes, dispensed into microwell strips precoated with H pylori cell membrane-derived antigen. The microwells were incubated at 37°C for one hour, washed in buffer, and 100ul antibody conjugate added, to be incubated again for 30 minutes and washed as above. Substrate chromogen (100ul of tetramethylbenzidine was added, shaken for 10 minutes, and the reaction was then stopped using 50ul of 2M sulphuric acid. Each microwell was then read at absorbance of 450nm in an ELISA reader (Labsystems, Uxbridge, UK). An antibody level of 10 units or more was considered as indicative of infection with H pylori.

To test for IgG H pylori antibodies by latex agglutination, Pyloriset (Orion Diagnostica, Finland) was used. The test uses latex particles coated with acid-extracted antigen of H pylori. A drop (40ul) of patient's serum, already diluted 1:2 in phosphate-saline buffer, was placed in the centre of the test slide, and one drop of latex reagent was added to be mixed with the serum. The test slide was then gently rotated for 3 minutes and observed for evidence of agglutination. A positive result was taken as white granules on a black background. In each series of tests positive and negative sera were used as controls.

Statistical analyses included Chi-square (X^2) test with correction for multiple comparisons, analysis of variance, and McNemar's test (124) where appropriate. Patients were endoscoped at random, and all specimens carried code numbers to facilitate randomization.

4.3 RESULTS

A total of 134 patients entered the final analysis: 75 took NSAID and 59 did not. Their characteristics are shown in Table 2.

Patients in both groups were comparable in their ages, smoking and drinking habits, and the occurrence of abdominal complaints. The details of NSAID and second-line drugs used by patients with rheumatoid arthritis (NSAID group) are presented in Table 3. NSAID taken by the majority of patients in this group are known for their potent anti-inflammatory activity and relatively marked ulcerogenic potential. The median duration of their intake was two years.

Patients not treated with NSAID, who are positive for H pylori according to various tests, are shown in Table 4. They are subgrouped according to the presence or absence of symptoms, and to their endoscopic and histological findings. Similar numbers of H pylori positive patients were identified by culture, histology, and CLO-test in various subgroups. Although such numbers were greater but not significantly different from those identified by the serological tests, the latter included more false positive cases which in turn resulted in lower specificity, as will be seen in Table 7.

H pylori positive patients in the NSAID group, classified according to their symptoms and endoscopic findings, are shown in Table 5. The serological tests, Helico-G in particular, diagnosed greater numbers of H pylori positive patients receiving NSAID but the significance of this finding is limited by the low specificity of

such tests (see below). It is also worth noting that patients treated with NSAID had a total of 30 cases of ulcers or erosions (30/75, 40%), and only 18 of these (18/30, 60%) were associated with abdominal complaints.

The histological abnormalities in patients receiving NSAID and the corresponding numbers of H pylori positive cases are presented in Table 6. The major abnormality was that of chronic superficial gastritis, followed by chemical gastritis. H pylori was identified in more than 60% of cases of chronic superficial gastritis by all tests used, with the greatest proportion (83%) diagnosed by Helico-G test. Six of 11 cases (54%) of chemical gastritis were associated with peptic damage, compared with 18 of 36 (50%) with H pylori, and 6 of 29 (21%, $p < 0.05$) in patients without chemical gastritis or H pylori.

The sensitivity, specificity, and the predictive values of various tests, compared with culture, are summarized in Table 7. Histology and CLO-test had the highest sensitivity, specificity, and predictive values, regardless of NSAID intake. Helico-G and Pyloriset tests had their lowest specificity and negative predictive values in patients treated with NSAID. Compared with Pyloriset, Helico-G had higher sensitivity and positive predictive values but lower specificity and negative predictive values, in the presence or absence of NSAID.

Using culture, histology and CLO-test, the prevalence of H pylori in the NSAID group was 43-48% compared with 64-69% in patients not treated with NSAID ($p < 0.01$). No significant differences were detected using Helico-G or Pyloriset tests. Also, H pylori IgG titres measured by Helico-G in patients taking NSAID were comparable

to those in patients not receiving such drugs. Of 15 patients treated with gold and NSAID 5 were positive for H pylori by culture, and this did not alter the interpretation of the above mentioned findings. The presence or absence of rheumatoid factor did not seem to influence the sensitivity or specificity of the serological tests used in this study.

TABLE 2 PATIENTS' DEMOGRAPHIC DETAILS

	<u>Patients</u>	<u>Patients</u>	
	<u>On NSAID</u>	<u>Not on NSAID</u>	<u>Total</u>
Number	75	59	134
Males	22	18	40
Females	53	41	94
Age (years)			
Median (interquartile			
ranges)	56 (44-66)	47 (36-65)	53 (39-65)
Smokers	27	21	48
Drinkers	34	33	67
Abdominal			
symptoms	36	39	75

TABLE 3. TYPES OF NSAID AND SECOND-LINE DRUGS USED

NSAID:

Indomethacin	17
Naproxen	9
Fenbufen	8
Diclofenac	7
Ketoprofen	6
Ibuprofen	6
Piroxicam	5
Flurbiprofen	5
Nabumetone	5
Others	6

Second-line Drugs:

Sulphasalazine	15
Sodium aurothiomalate	15
Hydroxychloroquine	7
Penicillamine	5

TABLE 4: H PYLORI POSITIVITY IN PATIENTS NOT TREATED WITH NSAID
(N=59), NO (%)

<u>Total number of patients</u>	<u>Patients positive for H pylori</u>				
	<u>Culture</u>	<u>Histology</u>	<u>CLO-test</u>	<u>Helico-G</u>	<u>Pyloriset</u>
Symptomatic patients (n=39)	32 (82%)	31 (80%)	31 (80%)	24 (62%)	24 (62%)
Endoscopic findings:					
- normal (n=53)	36 (68%)	35 (66%)	33 (62%)	34 (64%)	27 (51%)
- ulcers (n=6)	5	5	6	3	4
Histological findings:					
- normal (n=14)	5 (36%)	4 (29%)	5 (36%)	4 (29%)	3 (21%)
- chronic superficial gastritis (n=40)	33 (83%)	33 (83%)	32 (80%)	31 (78%)	27 (68%)
- chronic atrophic gastritis (n=3)	3	3	2	2	1

TABLE 5: H PYLORI POSITIVITY IN NSAID PATIENTS (N=75) WITH SYMPTOMS
AND/OR ENDOSCOPIC ABNORMALITIES. NO (%)

<u>Total number of patients</u>	<u>Patients Positive for H pylori</u>				
	<u>Culture</u>	<u>Histology</u>	<u>CLO-test</u>	<u>Helico-G</u>	<u>Pyloriset</u>
Symptomatic patients (n=36)	17(47%)	19(53)	16(44%)	24(67%)	19(53%)
Endoscopic findings:					
- Normal (n=45)	19(42%)	23(51%)	19(42%)	29(64%)	22(49%)
- Erosions (n=8)	3(38%)	3(38%)	3(38%)	5(63%)	4(50%)
- Gastric ulcers (n=14)	9(64%)	9(64%)	7(50%)	12(86%)	12(86%)
- Duodenal ulcers (n=8)	6(75%)	6(75)	5(63%)	5(63%)	3(38%)
Symptomatic endoscopic lesions (n=18)	9(50%)	10(56%)	8(44%)	12(67%)	11(61%)

TABLE 6: H PYLORI POSITIVITY IN NSAID PATIENTS (N=75)
WITH HISTOLOGICAL ABNORMALITIES. NO(%)

<u>Total number of patients</u>	<u>Patients Positive for H pylori</u>				
	<u>Culture</u>	<u>Histology</u>	<u>CLO-test</u>	<u>Helico-G</u>	<u>Pyloriset</u>
Chronic superficial gastritis (n=46)	30 (65%)	34 (74%)	29 (63%)	38 (83%)	30 (65%)
Lymphocytic gastritis (n=4)	4	4	4	2	3
Chemical gastritis (n=11)	1	1	1	5	5
Normal histology (n=14)	1	1	0	2	2

TABLE 7: SENSITIVITY, SPECIFICITY, POSITIVE (PPV) AND
 NEGATIVE (NPV) PREDICTIVE VALUES OF THE TESTS USED,
 WITH CULTURE TAKEN AS A STANDARD

	<u>Histology</u>	<u>CLO-test</u>	<u>Helico-G</u>	<u>Pyloriset</u>
<u>Patients not on NSAID</u>				
Sensitivity	95%	93%	75%*	63%*
Specificity	94%	94%	61%	67%
P P V	95%	93%	76%	61%
N P V	95%	95%	61%	67%
<u>Patients on NSAID</u>				
Sensitivity	97%	90%	81%	61%*
Specificity	90%	97%	45%*	50%*
P P V	97%	90%	83%	64%
N P V	90%	97%	44%	49%

Significant differences after correction for multiple comparison:

*:p < 0.05, by McNemar's test, compared with histology

4.3 DISCUSSION

This study shows that culture, histology and CLO-test have comparable sensitivity, specificity, positive and negative predictive values in diagnosing H pylori infection. Such characteristics do not seem to be influenced by treatment with NSAID. On the other hand, Helico-G and Pyloriset were less discriminatory, and the intake of NSAID was associated with lower specificity and negative predictive values with both of these serological tests.

Detecting H pylori in gastric biopsy specimens may be influenced by sampling errors (125). It is believed that this has been minimised by studying multiple histological sections made through each specimen and by assessing additional samples by culture and CLO-test as well as histology. The minimal differences in the sensitivity and specificity between these 3 methods can be explained by the presence of very few organisms in some biopsies, and this would be detected as a light growth by culture. On the other hand, the higher number of false positives diagnosed by the serology tests probably reflects previous infection with H pylori (126,127). The sensitivity of Helico-G found in this study is comparable to that of another report (120). However, it was not possible to confirm the sensitivity and specificity levels of Pyloriset, recently reported (121)

To the author's knowledge, all H pylori diagnostic tests have been assessed in patients not treated with NSAID. There is evidence to indicate that the natural history of peptic ulceration can be altered by such agents. Firstly, although more than 30% of NSAID takers might develop peptic damage (42,12), many such

lesions can be completely asymptomatic, as shown by this study and others (81). Secondly, NSAID seem to reduce the prevalence of H pylori infection, as suggested by data contained in this Thesis and elsewhere (23,75,83,84, Chapter 7). Such features of NSAID-induced peptic damage might limit the value of finding a positive IgG-related serological test in predicting ulcer disease in the presence or absence of abdominal complaints (79). This needs to be taken into consideration especially when serological testing is not supplemented with endoscopic, culture, or histological assessments (129).

The reason for the low prevalence of H pylori in the presence of NSAID remains a matter of speculation. It could be due to a direct toxic action against the organisms, or indirectly related to the effect of NSAID on the gastric mucosa. Aspirin and indomethacin were found to increase basal (13) and maximally stimulated gastric acid secretion (14,15). Aspirin (16) and indomethacin (17) were also shown to inhibit mucus secretion. This rise in gastric acid and the interference with the mucus layer by NSAID might in turn make it difficult for H pylori organisms to survive under such unfavourable conditions.

The median duration of NSAID intake in this study was two years. Assuming that some patients became negative for H pylori during such period, the behaviour of their serum antibody titres could not be predicted as the precise time of their conversion was not known. Recent studies have suggested that such titres might fall (130) or remain unchanged (126).

In this study, chemical gastritis was only found in patients receiving NSAID. H pylori IgG antibodies were detected in patients with chemical gastritis (5/11,45%) while the presence of H pylori organisms was confirmed in only one of these cases. This discrepancy between the serological and biopsy tests might again support the suggestion that the positive serology reflects previous infection with H pylori which could have been eradicated, or at least altered, by NSAID intake (23,75,83,84, Chapter 7).

NSAID related peptic ulcers or erosions were found in 54% of patients with chemical gastritis and 50% of those with H pylori, compared with only 21% of other patients in the NSAID group. This is in agreement with other findings in this Thesis suggesting that the prevalence of NSAID-induced peptic damage appears to be increased by the presence of chemical gastritis or H pylori (23, Chapter 7).

In conclusion, culture, histology and CLO-test have comparable sensitivity and specificity that do not seem to be influenced by NSAID intake. However, IgG antibody serology tests (Helico-G and Pyloriset) appear to be less useful than the above tests involving the use of gastric biopsy specimens. Such differences are further emphasised in patients treated with NSAID. The use of improved serological tests should still be considered because of their non-invasive nature, low cost, time and labour saving potential.

CHAPTER 5

5 EFFECT OF NSAID ON THE SERODIAGNOSIS OF H PYLORI

5.1 SUMMARY

Background: The host's humoral immune response to *Helicobacter pylori* has been utilized in the diagnosis of active infection with these organisms. Several commercial tests have become available, but the assessment of their efficacy has relied on few and unconfirmed reports, and in the absence of NSAID.

Aims: To assess and compare the efficacy of the following *H pylori* serological tests in patients treated or not treated with NSAID: Biolab Malakit, Bio-Rad GAP test IgG, Pyloriset Latex, and Helico-G tests.

Methods: Venous blood was tested at random in 124 patients: 64 received NSAID and 60 did not. *H pylori* IgG antibodies were detected by Latex agglutination (Pyloriset), or by ELISA (the remaining tests). Endoscopic gastric antral biopsies were also obtained for urease-activity, culture, and histology: detection of *H pylori* by at least two of them was considered as a true positive, and its absence in all biopsies as a true negative.

Results: The sensitivity values in the presence (or absence) of

NSAID were: Pyloriset Latex, 59% (60%); Helico-G, 79% (74%); Biolab Malakit, 85% (81%); and Bio-Rad GAP Test IgG, 100% (95%). The respective specificity values were: 50% (71%), 47% (59%), 50% (65%), and 30% (29%).

Conclusion: Bio-Rad GAP test IgG has the highest sensitivity and the lowest specificity values, regardless of NSAID intake. However, the sensitivity of the other tests is less than that of the standard biopsy-related tests and their specificity is even lower in chronic NSAID users.

5.2 INTRODUCTION

It is increasingly realised that the systemic immune response to *Helicobacter (H) pylori* infection confers no protection against the organisms, and its presence is of diagnostic value only (131). Several methods have been used to detect such responses including agglutination, complement fixation, and enzyme linked immunosorbent assay (ELISA) (116,119,121,132-135, Chapter 4). The low cost and convenience of these techniques encouraged some workers to recommend their use in the diagnosis and management of *H pylori* associated ulcers, without the need for endoscopy (79,80). Because of their diagnostic and therapeutic significance, increasing numbers of commercial serological tests are being introduced. The validity of such tests has, however, been assessed by few and unconfirmed studies, in the absence of NSAID, and direct comparison between them has been lacking. Recent evidence indicates that the prevalence of NSAID related ulcers is higher in patients infected with *H pylori* (23,75,129, Chapter 7), which emphasizes the need for

accurate diagnosis of this infection in such patients, and that the specificity of some serological tests can be influenced by NSAID (78, Chapter 4).

The aim of this study is to evaluate the efficacy of four newly introduced commercial tests (Pyloriset Latex, Helico-G, Biolab Malakit, and Bio-Rad GAP Test IgG) in diagnosing *H pylori* in patients with or without a history of chronic NSAID intake.

5.3 PATIENTS AND METHODS

Patients with adult rheumatoid arthritis (the NSAID group) were recruited from the Rheumatology Out-Patient Clinic provided they had taken their NSAID for at least four weeks. The rest of the patients were recruited from the Gastroenterology Out-Patient Clinic, where they had presented with transient or persistent abdominal complaints. They were excluded if they had been treated with steroids, cytotoxic drugs, antibiotics, anti-ulcer drugs, or if they had a history of gastro-duodenal surgery.

Venous blood, 10ml, was taken for serology tests and patients were endoscoped at random using 4-6mg midazolam for sedation, after giving an informed consent. A total of four gastric antral biopsies were taken for histology (two specimens), culture (one specimen), and CLO-test (one specimen). Ulcers were also biopsied to exclude malignancy.

H pylori was identified by culture and histology as previously described (61). The classification of gastritis was according to the

Whitehead system modified to include the newly described entities of lymphocytic and chemical gastritis (23,72,123, Chapter 7).

The CLO-test (Delta-West Ltd., Bentley, Western Australia) was carried out by placing the antral biopsy specimen into the gel pellet, and reviewing the slide for the positive red coloration at 3 and 24 hours (78, Chapter 4).

The Pyloriset Latex (Orion Diagnostica Finland) and the Helico-G (Porton Cambridge, Maidenhead, UK) tests were carried out according to the manufacturer's instructions and as recently described (78, Chapter 4). The Pyloriset detects IgG antibodies by agglutination, using Latex particles coated with acid-extracted antigen of *H pylori*. The Helico-G test uses ELISA to measure IgG antibodies directed against *H pylori* cell membrane-derived antigen.

ELISA was also used to detect IgG antibodies against two more partially purified antigens: *H pylori* urease antigen, by Biolab Mlakit (Biolab, Limal, Belgium), and *H pylori* outer membrane antigen, by Bio-Rad GAP test IgG, (Bio-Rad Chemical Division, Richmond, CA, USA). Both tests were also carried out according to the manufacturers' instructions. The Biolab Malakit test involved incubating the diluted samples and controls in microwells precoated with *H pylori* antigens at 37°C for 45 minutes, to form an *H pylori* antigen-antibody complex. The unbound components were washed off. An anti-IgG, coupled peroxidase conjugate, was then added and incubated, the unbound conjugate washed off, a substrate added and also incubated, and the reaction was finally stopped with NaOH. the absorbance was read at 405nm, with the intensity of the green

coloration being proportional to the amount of anti-H pylori antibodies in the sample. A sample was considered positive for H pylori if its optical density was higher than the limit of normal value, specified by the manufacturer. In Bio-Rad GAP test, 100ul of standards, controls and diluted patient samples were pipetted into the relevant microwells, and incubated for 60 minutes at 25°C. The contents were then discarded, the wells washed with the wash buffer solution and blotted on a paper towel. Anti-IgG enzyme conjugate solution was added after blue colour development, and absorbance was read at 450nm. A standard curve was constructed, and a sample was considered positive for H pylori if its antibody concentration was greater than 17 units.

A true positive was defined as a patient who had H pylori identified by at least two of the biopsy-related tests (histology, culture, and CLO-test), while a true negative was one who did not have H pylori diagnosed by any of such tests. Patients were excluded when H pylori was found by only one of the three biopsy-related tests.

Sensitivity was defined as the frequency of a positive test in all subjects with H pylori infection, and specificity was the frequency of a negative test in those without the infection.

Statistical analyses involved the Chi-square (X^2) and McNemar's test (124) with correction for multiple comparisons, where appropriate. All specimens carried code numbers, and were assessed under randomised conditions. The study was approved by the local Ethics Committee.

5.4 RESULTS

Four of 128 patients studied were excluded because H pylori was identified in their histological specimens only, and could not be confirmed by culture or CLO-test. The remaining 124 patients entered the final analysis: 64 took NSAID and 60 did not.

The demographic details of patients are shown in Table 8. Both groups of patients were comparable in their age, smoking, and drinking habits, but patients not treated with NSAID were more likely to have abdominal complaints. The median duration of NSAID intake was 2.5 years.

Table 9 demonstrates the prevalence of H pylori, as detected by the various tests, in patients with histological and endoscopic abnormalities. Chronic superficial gastritis was the commonest histological abnormality regardless of NSAID intake: similar numbers of patients with this abnormality, who were also positive for H pylori, were identified by Helico-G and Biolab Malakit as compared with the standard (or biopsy-related) tests. However, Pyloriset Latex identified the lowest number and Bio-Rad GAP test diagnosed the highest number of patients with both H pylori and chronic superficial gastritis. Chemical gastritis was found more commonly in patients receiving NSAID: H pylori organisms were seen in only one of nine patients, using the standard tests, but there were 5-6 seropositive cases with this histological entity. Also, a total of 19 patients treated with NSAID (29%) were found to have peptic ulcers, compared with seven (11%) in the absence of NSAID. Erosions were only found in patients taking NSAID. Almost all ulcers and erosions

were positive for H pylori by the serological tests.

The overall efficacy of the serological tests, in diagnosing H pylori infection, is shown in Table 10 and Figures 1 and 2. Helico-G and Biolab Malakit had a similar efficacy. However, Bio-Rad GAP test had the highest number of true and false positives, and as a result, the highest sensitivity and the lowest specificity regardless of NSAID intake. The specificity of Pyloriset Latex, Helico-G, and Biolab Malakit was lower in the presence of NSAID. Pyloriset Latex had the lowest sensitivity and highest specificity in patients not taking NSAID.

It is worth noting that the prevalence of H pylori in patients treated with NSAID was 34/64, 53%, compared with 43/60, 72% ($p < 0.05$), in patients not taking NSAID, using the standard tests, as shown in Table 10. The differences did not reach statistical significance when the findings of the serological tests were compared in the presence or absence of NSAID.

H pylori was identified in the gastric biopsies of six out of 17 rheumatoid patients (35%) receiving long-term gold injections plus NSAID, which is only two subjects less than the expected prevalence in such subgroup had they taken NSAID only: this did not alter the overall statistical analyses of the study.

TABLE 8
PATIENTS' GENERAL CHARACTERISTICS

	On NSAID	Not on NSAID
Number	64	60
Males	18	19
Females	46	41
Age (years) median	55	50
(interquartile range)	(43-65)	(38-65)
Smokers	25	21
Drinkers	31	34
Abdominal symptoms	25	39

TABLE 9: PATIENTS POSITIVE FOR H PYLORI CLASSIFIED ACCORDING TO
THEIR HISTOLOGICAL AND ENDOSCOPIC FINDINGS; NSAID

	Total No. patients	PRESENT (NSAID ABSENT)						Bio-Rad GAP Test IgG
		Standard tests	Pyloriset Latex	Helico-G	Biolab Malakit	Patients positive for H pylori		
Normal histology	19(12)	1(5)	2(3)	1(5)	1(5)	6(8)		
Chronic superficial gastritis	41(40)	30(34)	24(27)	34(31)	36(32)	39(38)		
Chemical gastritis	9(2)	1(0)	5(0)	5(0)	5(0)	6(1)		
Lymphocytic gastritis	4(3)	4(3)	3(3)	2(3)	3(3)	4(3)		
Chronic atrophic gastritis	0(3)	0(3)	0(1)	0(2)	0(2)	0(3)		
Gastric ulcers	12(2)	7(1)	10(0)	10(0)	10(0)	12(2)		
Duodenal ulcers	7(5)	6(5)	2(4)	4(3)	5(4)	7(5)		
Erosions	5(0)	2(0)	3(0)	4(0)	4(0)	4(0)		

TABLE 10: DETECTION OF H PYLORI BY SEROLOGY COMPARED WITH BIOPSY RELATED TESTS (STANDARD)

	Standard tests	Pyloriset		Helico-G		Biolab		Bio-Rad	
		Latex			Malakit		GAP Test	Igg	
On NSAID									
True positives	34	20	27	29	34				
True negatives	30	15	14	15	9				
False positives	-	15	16	15	21				
False negatives	-	14	7	5	-				
Not on NSAID									
True positives	43	26	32	35	41				
True negatives	17	12	10	11	5				
False positives	-	5	8	6	12				
False negatives	-	17	10	8	2				

Legends to Figures 1 and 2

The sensitivity (Figure 1) and specificity (Figure 2) of the serological tests in the presence or absence of NSAID.

*: $p < 0.05$ compared with the standard biopsy-related tests.

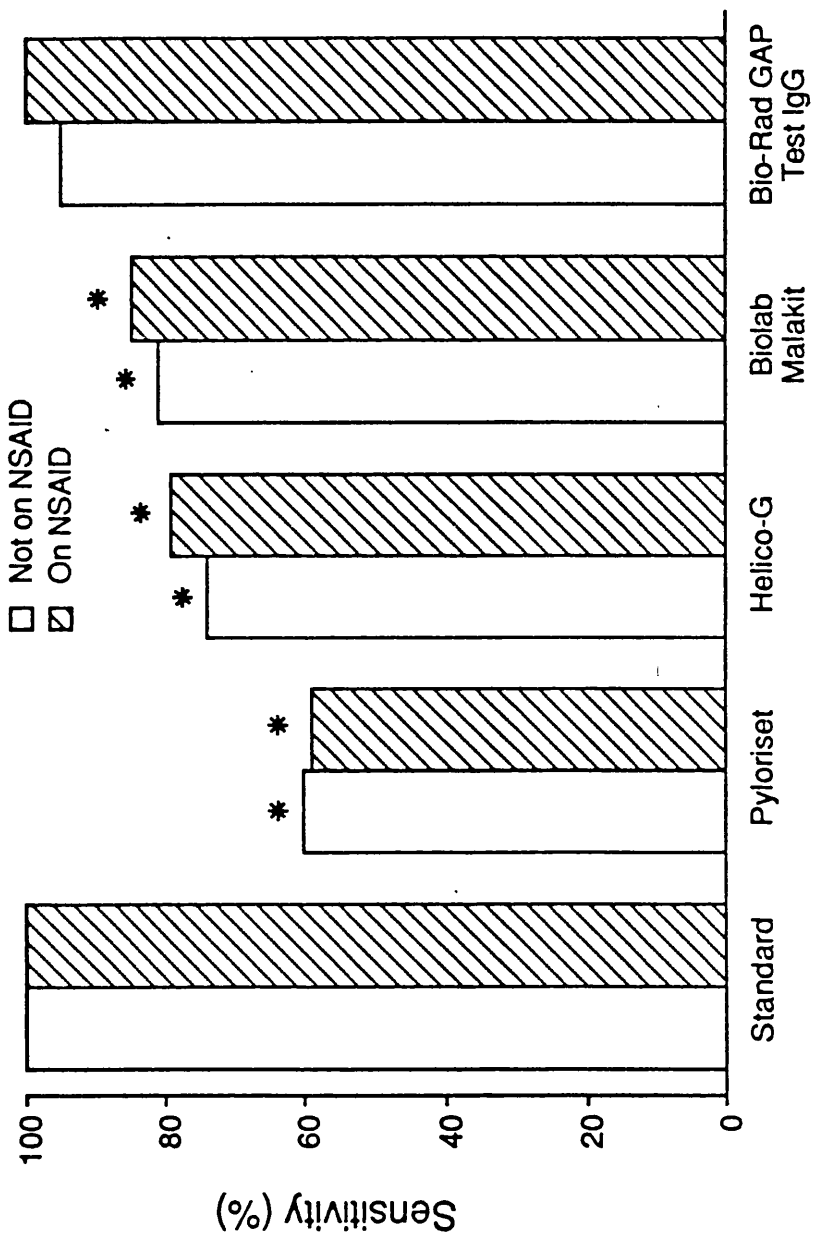


Figure 1

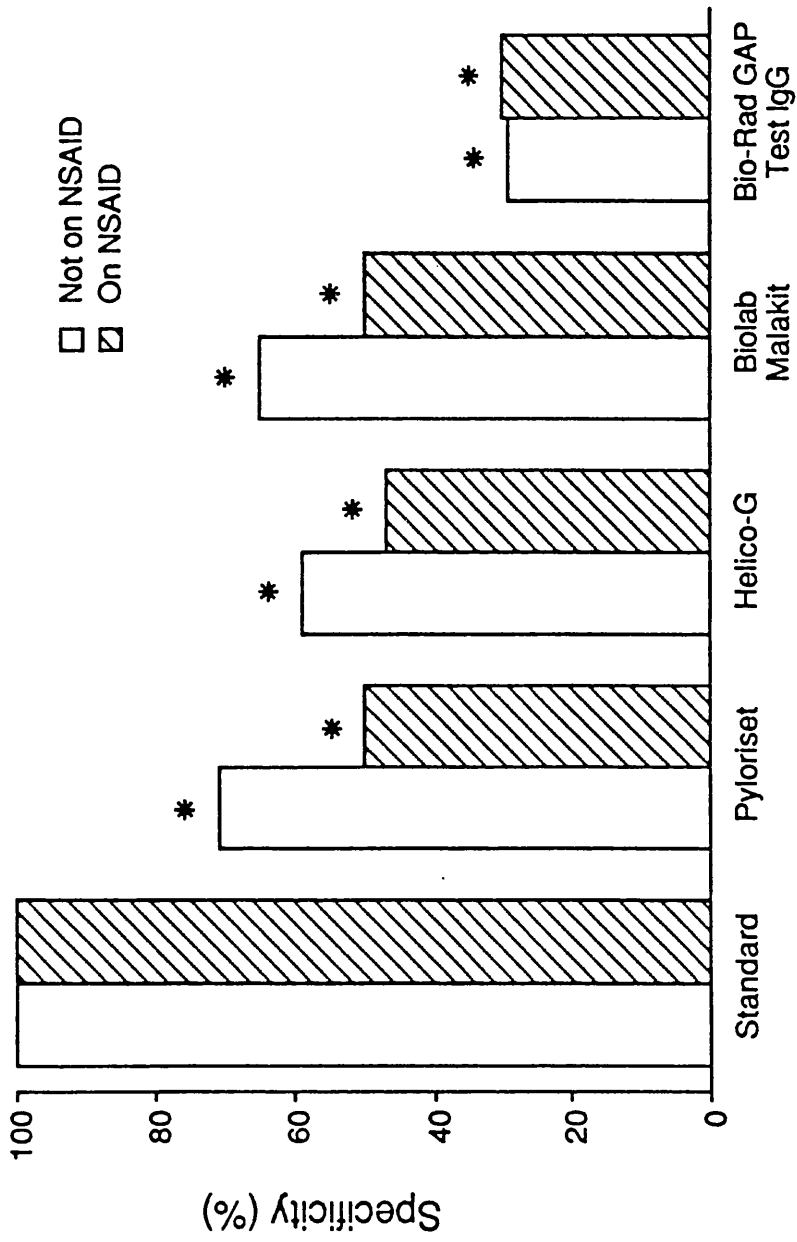


Figure 2

5.5 DISCUSSION

This study shows that the sensitivity and specificity of three commercial serological tests (Pyloriset Latex, Helico-G, and Biolab Malakit) are lower than those of the standard biopsy-related tests (culture, histology and CLO-test). The specificity values of the above serological tests are also lower in patients receiving long-term NSAID. The sensitivity of a 4th serological test, Bio-Rad GAP test IgG, is identical to that of the standard tests but its specificity is significantly lower.

Several factors have recently been identified as being capable of influencing the performance of H pylori serological tests: these include patients' age, their histological findings, H pylori antigens, and, as suggested by this study, NSAID intake.

H pylori infection and seropositivity have been shown to increase with age (96,136) although the immunological responses, in general, might be reduced in the elderly (137). Age is unlikely to have influenced the findings or the conclusions of this study, because all tests and comparisons were carried out on the same group of patients.

Patients with certain histological abnormalities might be seropositive for H pylori despite the failure to identify the organisms in their gastric biopsies. This is particularly true for cases of chemical gastritis, as shown by this study and by others (135), and might explain, at least in part, the reduced specificity of serology in NSAID patients, who have a higher prevalence of chemical gastritis

(13,22). Similar findings have also been reported in patients with chronic atrophic gastritis (138,139) which suggests that they have been infected with *H pylori* at some point in their lives. Atrophic gastritis was found in three patients in this study and is, therefore, unlikely to have influenced its results.

The development of serological assays for antibody responses to *H pylori* antigens has been complicated by the fact that *H pylori* organisms share antigens with other bacterial species, such as *Campylobacter jejuni* (140,141). A relatively high frequency of false-positive and false-negative results has been observed in studies using whole extracts of *H pylori* as antigens (87,132,136,140,141). More recently, higher sensitivity and specificity values have been reported when antibodies were raised against certain components of *H pylori* organisms such as partially purified urease enzyme or high molecular weight cell-associated proteins (119), and the 25-kilodalton antigen of *H pylori* (142). Another factor that needs to be considered is the fate of the antibody titres once their formation has been triggered by one or more of *H pylori* antigens. It has been suggested that a considerable number of healthy people previously infected with *H pylori* might remain seropositive despite the spontaneous elimination of the organisms (126). This supports the hypothesis that *H pylori* colonisation is a dynamic process with an active phase of infection and subsequent elimination of the bacteria in at least a proportion of infected people (126). Long-term serological surveillance studies have also found that in most patients without any detectable organisms, 12 months after anti-*H pylori* therapy, both specific IgA and specific IgG antibody values were lower but had not yet returned to the normal range (130). These

observations, like chemical gastritis, might also help understand the low specificity of the serological tests in this study especially in patients treated with NSAID, who have a low prevalence of H pylori.

Very few studies have been published on the evaluation of the serological tests described in this Chapter. Despite being directed against relatively specific components of H pylori, their sensitivity excluding that of Bio-Rad GAP Test, and their specificity values are lower than some of the recently published data. The findings with respect to Pyloriset Latex are similar to those of another report (143), but lower than the values described by another study (121), for unexplained reasons. The sensitivity and specificity of Helico-G in this study are also comparable to those of another (120), but it is worth noting that some workers (79) had to modify the manufacturer's instructions in order to improve the performance of Helico-G test. Some authors (119) have reported high sensitivity and specificity values for an ELISA test which, like Biolab Malakit, detects antibodies against partially purified urease antigen. However, the same test (119) has been used by another group of workers (139) who found that 25 out of 124 patients studied (20%) were seropositive for H pylori despite the absence of the organisms in their gastric biopsy specimens; 20 of the 25 false positive cases had atrophic gastritis (139).

The sensitivity of Bio-Rad GAP test IgG described in this study is almost identical to that found by another group (135), but the lower specificity could be explained by one or more of the following: their reliance on histology as a standard, which might be difficult to interpret in the presence of very few organisms; the exclusion from their study of patients with chronic superficial or atrophic gastritis

in whom *H pylori* was not identified, thus reducing the number of true negatives; the relatively high number of cases of chemical gastritis in their *H pylori* negative group without specifying whether such patients had been treated with NSAID; and the presence in their *H pylori* positive group, of more patients with chronic atrophic gastritis (117 out of 160, 73%), which might have been paralleled by a rise in the number of false positive cases (138,139).

The lower specificity of Pyloriset Latex, Helico-G, and Biolab Malakit in NSAID patients could be explained by the presence of chemical gastritis (23,71,72,135, Chapters 4,7) and the low prevalence of *H pylori* in such patients, as found by this study and others (23,83,85). The latter could be related to the impairment, by NSAID, of the gastric mucus layer, which is considered as the natural habitat of *H pylori* organisms (23). However, when these bacteria persist in the presence of NSAID, it has been suggested that the damaging effects of NSAID might become intensified (23,129, Chapter 7). Therefore, accurate diagnosis of *H pylori* in such patients is, at least, as important as it is in patients not taking NSAID.

In conclusion, this study has evaluated the efficacy of four commercial serological tests in comparison with standard biopsy-related tests. Despite having high sensitivity, Bio-Rad GAP Test IgG has been found to have low specificity. Pyloriset Latex, Helico-G, and Biolab Malakit all had relatively low sensitivity and specificity, the latter being even lower in the presence of NSAID. These results could be related to the histological findings, the dynamic nature of *H pylori* infection, the types of the antigens

tested, NSAID intake, or due to some weaknesses in the manufacturers' instructions. These factors have to be considered in order to improve the reliability of serodiagnosis because of the potential advantages of such methods in identifying active H pylori infection.

CHAPTER 6

6. PREDICTION OF NSAID-RELATED PEPTIC ULCERS USING THE HEALTH ASSESSMENT QUESTIONNAIRE

6.1 SUMMARY

Background: Patients with disabling manifestations of arthritis might be at a greater risk of developing NSAID-related peptic ulcers, as their advanced disease might require more aggressive therapy.

Aims: To assess the possibility of a relationship between the degree of physical disability in arthritic patients and non-steroidal peptic ulceration.

Methods: Patients were endoscoped immediately after performing their arthritic functional assessments. The Health Assessment Questionnaire was used and patients were classified into three main groups: I, scoring 0-1; II, 1.1-2; and III, 2.1-3.

Results: Ulcers, found in 36 out of 89 patients studied (36/89, 40%), were distributed as follows: 15/22 (68%) in group III compared with 9/28 (32%) in group I ($X^2=5.2$, $p<0.02$), and 12/39 (31%) in group II ($X^2=7.24$, $p<0.01$).

Conclusion: Patients with debilitating arthritis appear to have a higher prevalence of non-steroidal peptic ulceration. This finding might be relevant to the process of selecting patients for prophylactic anti-ulcer therapy.

6.2 INTRODUCTION

The link between peptic ulcers, or their complications, and non-steroidal anti-inflammatory drugs (NSAID) has been emphasized by several studies in recent years (42,95,144,145). Evidence of peptic damage can be found in more than 30% of patients receiving NSAID (42,128). The predisposing factors for NSAID-related peptic ulcer are not fully understood, and it is not known whether the likelihood of developing such ulcers is influenced by the degree of physical disability in arthritic patients. The Health Assessment Questionnaire (H.A.Q) has been widely accepted as a useful tool for measuring the functional outcome of arthritis (146-148). Using H A Q and endoscopy, this study tested the possibility that the prevalence of peptic ulcers varies amongst different functional groups of arthritic patients being treated with NSAID.

6.3 PATIENTS AND METHODS

Patients with osteoarthritis or rheumatoid arthritis, diagnosed according to the criteria of the American Rheumatism Association (149) were recruited provided they were 18 years or over, and took NSAID for a minimum of 4 weeks. Second-line drugs were allowed if they were started at least 6 months before the study. Patients were

excluded if they had previous gastric surgery or if they took ulcer healing drugs, steroids or cytotoxic agents.

The degree of physical disability was assessed using H A Q (146-148) (Figure 3) which in turn investigated patients' ability to dress themselves, rise, eat, walk, maintain their hygiene, reach, grip, perform other activities (shopping, getting in and out of a car, vacuuming and light gardening), and their need to use aids or supportive devices. Patients were classified into 3 main functional groups: group 1, capable of performing the above functions with some or no difficulty (scoring 0-1); group II, finding much difficulty (scoring 1.1-2); and group III, in need of several devices or unable to carry out the above functions (scoring 2.1-3). Patients' joints' pain was evaluated on a 4-point scale: none, mild, moderate and severe.

Informed consent was obtained and endoscopy was carried out within 2 hours of performing the functional assessment. The endoscopist was not aware of the functional scores. At endoscopy, sedation was achieved using 4-8mg midazolam intravenously. Biopsies were taken from the ulcer edge and base to exclude malignancy, and from the gastric antrum to check for *Helicobacter* (H) pylori by histology and bacteriology, as previously described (61). An ulcer was defined as a three-dimensional interruption in the mucosa measuring at least 3mm in diameter (41).

Statistical tests included the Chi-square (X^2) test, the Mann-Whitney, multivariate and logistical regression analyses, where appropriate.

The study was approved by the local Ethics Committee.

6.4 RESULTS

Eighty-nine patients were studied. They were classified as follows: 28 in group I, 39 in group II and 22 in group III. Their demographic details are shown in Table 11. Patients were comparable in their age, weight, and serum albumin levels. Weight and serum albumin were used as a rough estimate of patients nutritional status. There were no significant differences in the number of smokers amongst the various groups. Group I contained relatively more males and alcohol drinkers, while patients in group III were more likely to have abdominal complaints.

The duration of arthritis did not differ significantly amongst the study groups, as shown in Table 12. Group III patients tended to have severe joint pain; they also had higher erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP). Similar NSAID and second-line agents were taken by comparable numbers of patients in all groups (Tables 12 and 13). The doses and duration of NSAID used were also similar.

The prevalence of peptic ulcers was higher in group III (Table 14), but the prevalence of H pylori and the histological findings were similar amongst the various groups.

Peptic ulcers described in this study were found in the gastric antrum and the first part of the duodenum. They were distributed as follows 5 gastric (4 duodenal) in group I, 8 (4) in group II, and 10 (5) in group III.

The differences in the prevalence of peptic ulcers remained significant even after allowing for minor variations in the demographic characteristics of patients, as tested by the multivariate and logistical regression analyses.

FIGURE 3

The nature of the Health Assessment Questionnaire. It is specially designed for use in osteoarthritis and rheumatoid arthritis, and provides an overall "Disability Index" from 0 (no disability) - 3 (complete dependence on others). It may be used to estimate the impact of arthritis on daily living. It may also help to assess changes in disease activity over several months, or to monitor disease progression year by year.

The overall score for the index is the mean of the 8 numbered sections taken together. The score for each section is that of the worse response to any of the questions in that section. The score for each response is: without any difficulty = 0, with some difficulty = 1, with much difficulty = 2, and unable to do = 3. If appliances or devices or help from another person is required for activities within a section (indicated at the bottom of each page), then that section scores at least 2 (it may score 3 if the patient's response was "unable to do").

HEALTH ASSESSMENT QUESTIONNAIRE

Name Date

We are interested in learning how your illness affects your ability to function in daily life.
Please feel free to add any comments at the end of this form.

PLEASE TICK THE ONE RESPONSE WHICH BEST DESCRIBES YOUR USUAL ABILITIES
OVER THE PAST WEEK:

	Without ANY difficulty	With SOME difficulty	With MUCH difficulty	Unable to do
1. DRESSING AND GROOMING				
Are you able to:				
- Dress yourself, including tying shoelaces and doing buttons?
- Shampoo your hair?
2. RISING				
Are you able to:				
- Stand up from an armless straight chair?
- Get in and out of bed?
3. EATING				
Are you able to:				
- Cut your meat?
- Lift a full cup or glass to your mouth?
- Open a new carton of milk (or soap powder)?
4. WALKING				
Are you able to:				
- Walk outdoors on flat ground?
- Climb up five steps?

PLEASE TICK ANY AIDS OR DEVICES THAT YOU USUALLY USE FOR ANY OF THESE ACTIVITIES:

- | | |
|-----------------------------|--|
| Cane | Devices used for dressing (button hook, zipper pull, long handled shoe horn, etc.) |
| Walking frame | Built-up or special utensils |
| Crutches | Special or built-up chair |
| Wheelchair | |
| Other (specify) | |

PLEASE TICK ANY CATEGORIES FOR WHICH YOU USUALLY NEED HELP FROM ANOTHER PERSON:

- | | |
|-----------------------------|---------------|
| Dressing and Grooming | Eating |
| Rising | Walking |

PLEASE TICK THE ONE RESPONSE WHICH BEST DESCRIBES YOUR USUAL ABILITIES OVER THE PAST WEEK

	Without ANY difficulty	With SOME difficulty	With MUCH difficulty	Unable to do
5. HYGIENE				
Are you able to:				
- Wash and dry your entire body?
- Take a bath?
- Get on and off the toilet?
6. REACH				
Are you able to:				
- Reach and get down a 5lb object (e.g. a bag of potatoes) from just above your head?
- Bend down to pick up clothing from the floor?
7. GRIP				
Are you able to:				
- Open car doors?
- Open jars which have been previously opened?
- Turn taps on and off?
8. ACTIVITIES				
Are you able to:				
- Run errands and shop?
- Get in and out of a car?
- Do chores such as vacuuming, housework or light gardening?

PLEASE TICK ANY AIDS OR DEVICES THAT YOU USUALLY USE FOR ANY OF THESE ACTIVITIES:

- | | |
|---|---|
| Raised toilet seat | Bath rail |
| Bath seat | Long handled appliances for reach |
| Jar opener (for jars previously opened) | Other (specify) |

PLEASE TICK ANY CATEGORIES FOR WHICH YOU USUALLY NEED HELP FROM ANOTHER PERSON:

- | | |
|---------------|-----------------------------------|
| Hygiene | Gripping and opening things |
| Reach | Errands and housework |

TABLE 11: DEMOGRAPHIC CHARACTERISTICS OF PATIENTS IN VARIOUS FUNCTIONAL GROUPS. NO (%), MEDIAN (INTERQUARTILE RANGES)

	<u>Group I (n=28)</u>	<u>Group II (n=38)</u>	<u>Group III (n=22)</u>
Males	13 (46%)	6 (15%)	4 (18%)
Females	15 (54%)	33 (85%)	18 (82%)
Age (years)	55 (40-60)	55 (45-66)	58 (51-60)
Osteoarthritis	6 (21%)	7 (18%)	2 (9%)
Rheumatoid arthritis	22 (79%)	32 (82%)	20 (91%)
Smoking	11 (39%)	12 (31%)	10 (45%)
Drinking	16 (64%)	18 (46%)	7 (31%)
Abdominal symptoms	10 (36%)	10 (26%)	11 (50%)
Weight (kg)	66 (55-82)	62 (56-69)	64 (52-75)
Serum albumin (g/l)	42 (41-45)	42 (38-45)	41 (39-44)

TABLE 12: DURATION OF ARTHRITIS, ITS ACTIVITY, AND THE USE OF SECOND LINE AGENTS. NO (%), MEDIAN (INTERQUARTILE RANGES)

	<u>Group I (n=28)</u>	<u>Group II (n=38)</u>	<u>Group III (n=22)</u>
Duration of arthritis (yrs)	6 (3-10)	8 (4-12)	9 (5-14)
Joint pain -			
Mild	17 (61%)	16 (41%)	4 (18%)
Moderate	8 (29%)	19 (49%)	8 (36%)
Severe	3 (11%)	4 (10%)	10 (45%)
Erythrocyte Sedimentation Rate (mm/hr)	16 (6-27)	30* (14-49)	32** (22-60)
C-Reactive protein mg/l	12 (10-28)	12 (10-30)	22*** (12-56)
Second line drugs-			
Gold	5 (18%)	6 (15%)	8 (36%)
Sulphasalazine	7 (25%)	9 (23%)	4 (18%)
Penicillamine	1 (4%)	3 (8%)	1 (5%)
Hydroxychloroquine	5 (18%)	5 (13%)	-

Significant rise: * p,0.05, ** p<0.01 (Vs.I)

*** p<0.05 (Vs I,II)

TABLE 13: DETAILS OF NON-STEROIDAL ANTI-INFLAMMATORY DRUGS
 NO. (%), MEDIAN (INTERQUARTILE RANGES)

<u>TYPE</u>	<u>Group I (n=28)</u>	<u>Group II (n=38)</u>	<u>Group III (n=22)</u>
Naproxen	7 (25%)	7 (18%)	3 (14%)
Indomethacin	4 (14%)	8 (21%)	7 (32%)
Diclofenac	4 (14%)	5 (13%)	3 (14%)
Ketoprofen	3 (11%)	2 (5%)	3 (14%)
Fenbufen	4 (14%)	4 (10%)	1 (5%)
Ibuprofen	1(4%)	3 (8%)	2 (9%)
Flurbiprofen	1(4%)	2 (5%)	2 (9%)
Others	4 (14%)	8 (21%)	1 (5%)
<u>Dosage*</u>	2 (2-3)	3(2-3)	2 (2-3)
<u>Duration (years)</u>	2 (1.5-4)	2 (1.5-3)	2 (1-5)

* 1: minimal

2: submaximal, and

3: maximal dose

TABLE 14: THE PREVALENCE OF PEPTIC ULCERS, H PYLORI, AND THE HISTOLOGICAL FINDINGS IN THE FUNCTIONAL GROUPS

	<u>Group I (n=28)</u>	<u>Group II (n=39)</u>	<u>Group III (n=22)</u>
Peptic ulcers	9 (32%)	12 (31%)	15 (68%)*,**
H pylori	12 (43%)	17 (44%)	11 (50%)
Gastric histology:			
- normal	10 (35%)	9 (23%)	6 (27%)
- chronic superficial gastritis	12 (43%)	21 (54%)	12 (55%)
- Chemical gastritis	4 (14%)	7 (18%)	4 (18%)
- Lymphocytic gastritis	2 (7%)	- -	- -
- Chronic atrophic gastritis	- -	2 (5%)	- -

Significant rise: * $X^2 = 5.2$, $p < 0.02$ (Vs.I)

** $X^2 = 7.24$, $p < 0.01$ (Vs.II)

6.5 DISCUSSION

This study suggests that arthritic patients with the greatest degree of functional disability, as measured by H A Q have the highest prevalence of peptic ulcers. Recent studies have indicated that NSAID-induced ulcers were found more frequently in smokers (42), in patients receiving stronger NSAID (150) and in those infected with H pylori (23,84, Chapter 7). Such ulcers were, however, less frequent in patients receiving gold therapy (82, Section VI). These observations are unlikely to explain the findings of this study: patients in the various functional groups were comparable in their age, smoking habits, NSAID, second-line drugs, and the prevalence of H pylori. Another factor that has recently received attention, because of its correlation with NSAID-related peptic ulceration, is that of chemical gastritis (23,71, Chapter 7). This has also occurred almost equally in the three functional groups. The similarity in patients' weights and serum albumin levels makes it unlikely for the results to have been influenced by any significant degree of malnourishment.

It could be argued that patients in group III had more ulcers because their NSAID intake might have been greater as a result of the higher activity of their arthritis. However, it is difficult to see how this would explain the similarity in the prevalence of ulcers in group I and II, who had different degrees of disease activity as suggested by the ESR and joint pain evaluations. Moreover, no significant differences were found in the doses or duration of NSAID taken by the various groups, as shown in Table 13. Alternatively, it could be speculated that the poor functional outcome of arthritis

might be associated with weakening of mucosal defences by stress, acid, disturbance in mucosal blood flow or prostaglandin production, or by as yet unknown mechanism. The relative immobility of group III patients could also be associated with poor gut motility and delayed gastric emptying which in turn might increase the risk of NSAID-induced ulcers. Such speculations will obviously need to be verified by further investigations.

In order to prevent NSAID-related peptic ulcers, prophylactic anti-ulcer therapy has been suggested for patients receiving such drugs (41). Concerns about the cost and side-effects of long-term anti-ulcer agents have lead to calls for targeting patients who would benefit most from such prophylactic treatment (151). It is believed that this process might be facilitated by the use of H A Q: arthritic patients with severe physical disability, as assessed by H A Q, have a higher prevalence of peptic ulceration.

SECTION IV
NSAID-RELATED HISTOLOGICAL CHANGES IN THE
UPPER GASTROINTESTINAL TRACT

INTRODUCTION

Very little is known about the effect of NSAID on the histology of the upper gastro-intestinal tract. The importance of this issue relates to its potential to clarify several aspects of the pathogenesis of NSAID-related damage.

Chapters included in Section IV study the histological changes in the stomach, duodenum, and oesophagus in the presence or absence of NSAID, in patients with or without local ulceration.

CHAPTER 7

7. NSAID AND THE GASTRIC MUCOSA

7.1 SUMMARY

Background: The stomach is the commonest site of both NSAID-related ulcers and H pylori infection. It is not known whether NSAID can cause a specific form of inflammation or "chemical gastritis", similar to that caused by other chemicals such as bile or alcohol.

Aims: To evaluate the prevalence and significance of chemical gastritis, in comparison with gastritis related to Helicobacter pylori in patients receiving NSAID.

Methods: Patients with or without a history of chronic NSAID intake were studied. Endoscopic biopsies were taken for histology and to test for H pylori. Chemical gastritis was defined as the presence of foveolar hyperplasia, vasodilatation, oedema, paucity of inflammatory cells, and the presence of muscle fibres in the lamina propria.

Results: 218 patients were studied: 174 took NSAID and 44 did not. Chemical gastritis was found in 46 patients (26%) in the NSAID group, and 3 (7%) in subjects not on these drugs ($p < 0.01$). H pylori was detected in 56 subjects (32%) on

NSAID compared with 22 (50%) in subjects not taking these agents ($p < 0.02$). In the NSAID group, ulcers were found in 16 out of 72 patients (16/72, 22%) without H pylori or chemical gastritis compared with 27/56 (48%) of patients with H pylori related gastritis ($p < 0.01$), and 25/46 (54%) of patients with chemical gastritis ($p < 0.001$).

Conclusion: NSAID related peptic ulcers appear to occur more commonly in patients with chemical gastritis or H pylori related gastritis. Also, patients on NSAID have a greater prevalence of chemical gastritis but lower prevalence of H pylori than those not taking these drugs.

7.2 INTRODUCTION

The effect of non-steroidal anti-inflammatory drugs (NSAID) on gastric histology is not fully clear. Recent studies (73,74) have reported some histological changes similar to those encountered in patients with previous gastric surgery (72). They consisted of foveolar hyperplasia, vasodilatation, oedema, lack of inflammatory cells, and the presence of muscle fibres in the lamina propria. These features were initially called "reflux gastritis", referring to the effect of bile reflux on the post-operative stomach (72). Since other chemicals, such as NSAID, were also thought to be involved in the intact stomach, the terms type C or "chemical" gastritis were proposed (71). The aim of this study was to evaluate the prevalence and significance of chemical gastritis, in comparison with gastritis related to Helicobacter (H) pylori, in patients taking NSAID. This could also clarify the interaction that might exist

between H pylori and NSAID in the gastric mucosa.

7.3 PATIENTS AND METHODS

Patients with adult onset rheumatoid arthritis were recruited from the Rheumatology Clinic provided they had taken NSAID for a minimum of 4 weeks, and excluded if they had received anti-ulcer therapy, antibiotics, or cytotoxic drugs. Patients not on NSAID were also accepted provided they had not taken these agents or any of the drugs mentioned above for at least a month. Subjects with a history of previous gastric surgery were excluded. All patients were asked about their alcohol intake. This was considered heavy if the subject took 10 or more units per week.

Informed consent was obtained and endoscopy performed after the administration of 5-15mg diazepam intravenously. The endoscopic findings were graded as follows: 0, normal or minor erythema; 1, 1-5 erosions; 2, 6-10 erosions; 3, frank ulceration measuring at least 5mm in diameter.

An average of 3 biopsy specimens were obtained from healthy looking mucosa in the gastric antrum of each subject. They were fixed in 10% formalin/saline and processed routinely. Cuts were made at 3 levels in the paraffin wax sections, which were then stained with haematoxylin and eosin. Further sections were also made and treated with cresyl violet to help identify H pylori.

The findings in each set of biopsies were graded twice: firstly, according to the Whitehead classification of gastritis (122) modified to

cover the newly described entity of lymphocytic gastritis (123), and secondly, according to a system modified from Dixon et al. for the diagnosis of chemical gastritis (72). Features of the modified Whitehead gastritis (Figures 4,5) included neutrophilic infiltration, chronic inflammatory cells, lymphocytic aggregates, intestinal metaplasia and gland atrophy. Their degree was graded on a four point scale: 0, absent; 3, severe. A similar scale was applied to the following features of chemical gastritis (Figure 6): (i) the degree of foveolar hyperplasia, (ii) the degree of oedema and the prominence of muscle fibres in the lamina propria, (iii) the degree of vasodilatation and congestion, (iv) the inverse of the number of neutrophils (absence scoring 3), and (v) the inverse of the number of plasma cells (absence again scoring 3). Patients scoring 8 or more by the chemical scores were considered to have chemical gastritis.

Two further biopsies were taken from the gastric antrum of each subject, and examined for the presence of *H pylori* by both histology and bacteriology, as previously described (61).

Statistical analyses: Chi-square (X^2) test with Bonferroni correction for multiple comparisons was used. Spearman's coefficient was also used to test for the correlation between the endoscopic and histological scores. The pathologist, and bacteriologist were unaware of patients' drugs. Code numbers were used to facilitate randomization of biopsy specimens.

FIGURE 4

Antrum of stomach: Chronic superficial gastritis, typical of *Helicobacter* related gastritis. It shows foci of active gastritis with degradation of the surface epithelium. HE X 235.

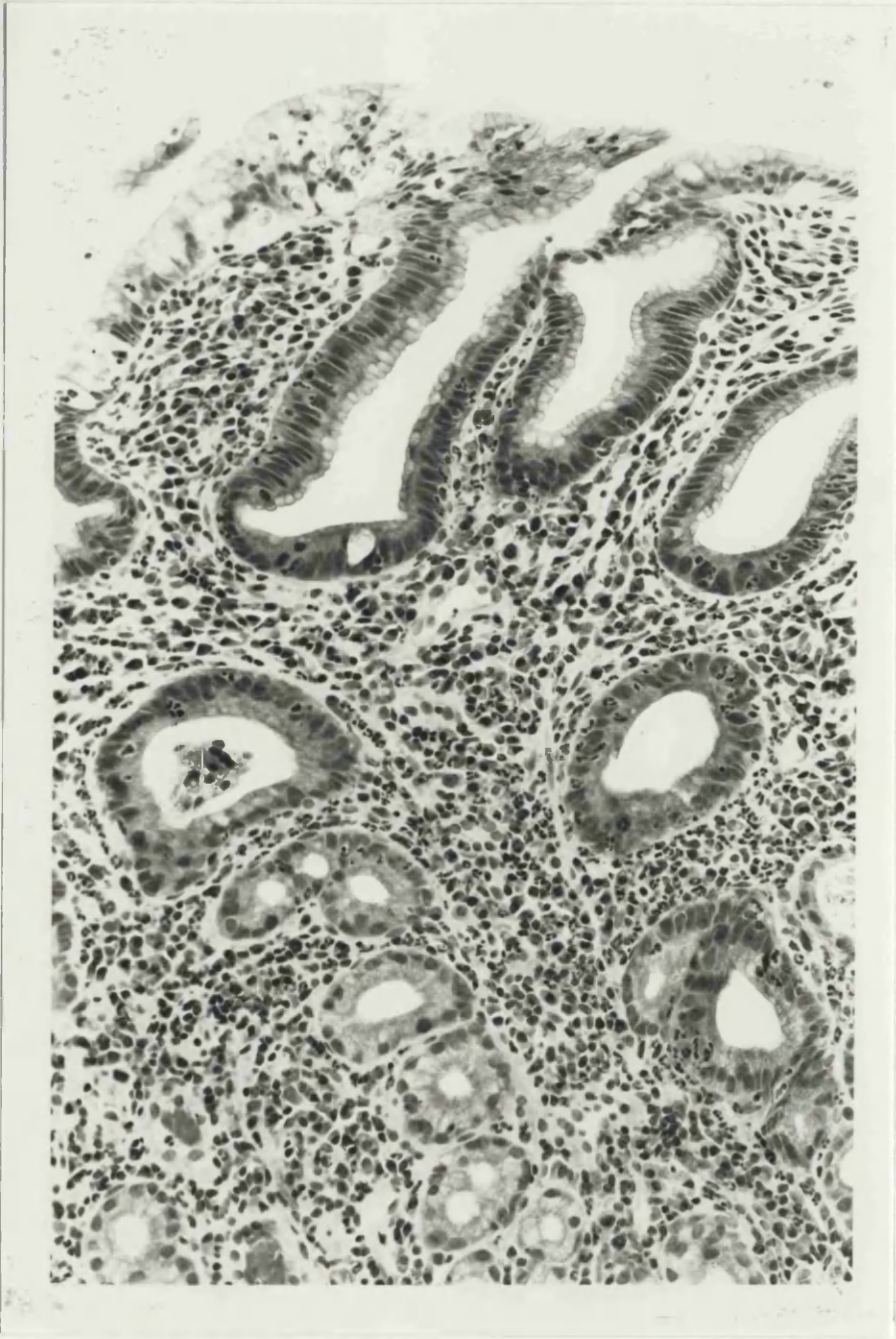


FIGURE 5

Antrum of stomach: Lymphocytic gastritis. There is a pronounced increase in lymphocytes in the surface and foveolar epithelium. HE X 314



FIGURE 6

Antrum of stomach: "Chemical" gastritis. Note the elongation of the foveolae, the virtual absence of inflammatory cells and the presence of oedema and muscle fibres in the foveolar compartment.

The "chemical score" was 10. HE X 170

7.4 RESULTS

A total of 210 patients were studied: 124 took NSAID, and 86 did not. Their characteristics are shown in Table 17. Patients not on



found in this group. There was no significant difference in the distribution of ulcers, or crypts or the histological findings between rheumatoid patients or those with other musculo-skeletal disorders not taking NSAID. The distribution of ulcers in NSAID histological

7.4 RESULTS

A total of 218 patients were studied: 174 took NSAID, and 44 did not. Their characteristics are shown in Table 15. Patients not on NSAID included 27 subjects with rheumatoid arthritis, and 17 with other musculo-skeletal disorders: 13 with osteoarthritis, and 4 with ankylosing spondylitis.

The prevalence of chemical gastritis and H pylori, in the presence or absence of NSAID, is shown in Figure 7. Subjects receiving NSAID were more likely to develop chemical gastritis, but less likely to have H pylori than those not on NSAID. Two of the three patients with evidence of chemical gastritis, in subjects not on NSAID, had a history of heavy alcohol intake. They both had rheumatoid arthritis.

Patients with chemical gastritis, H pylori, and those with neither of these findings in the NSAID group, tended to have similar demographic data, as shown in Table 16. H pylori could not be detected in any of the patients with chemical gastritis. Histological findings, other than chemical gastritis are presented in Table 17. Chronic superficial gastritis was the major finding in all groups.

Patients not on NSAID had a total of 14 ulcers: 5 gastric and 9 duodenal, all of which were positive for H pylori. No erosions were found in this group. There was no significant difference in the distribution of ulcers, H pylori or the histological findings between rheumatoid patients or those with other musculo-skeletal disorders not taking NSAID. The distribution of ulcers in NSAID histological

subgroups is presented in Figure 8; this shows that NSAID related peptic ulcers are more likely to develop in the presence of H pylori or chemical gastritis. However, the correlation between the endoscopic and histological scores was significant only in those with chemical gastritis, as shown in Table 18. Erosions were found in 11 NSAID patients without H pylori or chemical gastritis. Erosions in the other NSAID subgroups are shown in Table 18. A total of 68 ulcers were diagnosed in the NSAID group: 58 gastric, and 10 duodenal. Twenty of 58 NSAID related gastric ulcers (20/58,34%) and 5/10 (50%) duodenal ulcers were positive for H pylori.

TABLE 15: Characteristics of Patients with or without a History of NSAID Intake

	On NSAID	Not on NSAID
Number of patients	174	44
Age (years)	59	53
Median (Interquartile range)	(51-65)	(34-65)
Males	29	23
Females	145	21
Smokers	35	20
Drinkers:		
Less than 10 units/week	36	17
10 or more units/week	3	8
Upper abdominal symptoms	63	42

TABLE 16: NSAID Patients Subgrouped According to their Main Histological Findings

	Chemical gastritis	H pylori related gastritis	No H pylori No chemical gastritis
Number of patients	46	56	72
Age (years)	60	57	62
Median (interquartile range)	(52-69)	(50-65)	(57-65)
Males	6	9	14
Females	40	47	58
Smokers	7	11	17
Drinkers:			
less than 10 units/week	10	11	15
10 or more units/week	0	2	1
Upper abdominal symptoms	15	19	29
Second-line agents:			
Gold	6	5	8
Sulphasalazine	6	3	6
Penicillamine	3	3	8
Hydroxychloroquine	3	2	5

TABLE 17: Histological Findings in Patients Without Chemical Gastritis

	On NSAID				Total	Not on NSAID
	No H pylori		No chemical			
	H pylori Present	No H pylori	No chemical	Gastritis		
Number of patients	56	72	128	41		
Normal	0	16	16	7		
Chronic superficial gastritis	48	44	92	31		
Chronic atrophic gastritis	5	10	15	1		
Chronic lymphocytic gastritis	3	2	5	2		




TABLE 18: Correlation between Endoscopic and Histological Scores
in NSAID Patients with Chemical Gastritis or H pylori

Histological Scores	Endoscopic Scores			
	0	1	2	3
Chemical:**				
8	8	4	2	3
9	2	0	1	10
10	2	1	1	10
11	0	0	0	2
H pylori-related:**				
3-5	9	6	0	12
6-8	8	3	1	11
9	1	1	0	4

*: $r = 0.51$, $p < 0.05$ **: $r = 0.0023$, not significant

FIGURE 7

**The prevalence of H pylori and chemical gastritis in the presence or
absence of NSAID**

- A  Total no. of subjects
- B  Subjects with H.pylori
- C  Subjects with chemical gastritis

* $X^2 = 7.95$ $p < 0.01$ (C₁ vs. C₂)
 ** $X^2 = 4.43$ $p < 0.02$ (B₂ vs. B₁)

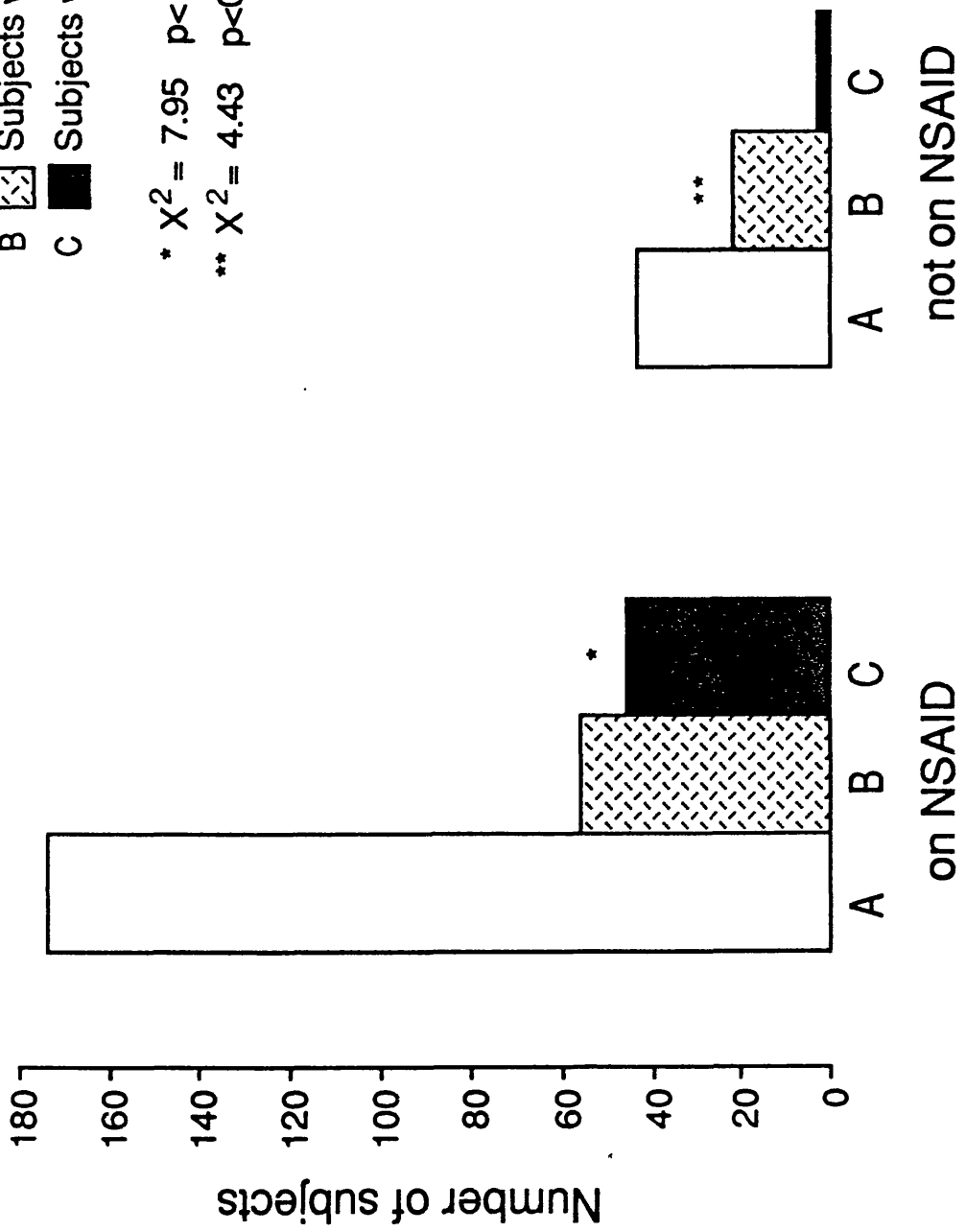
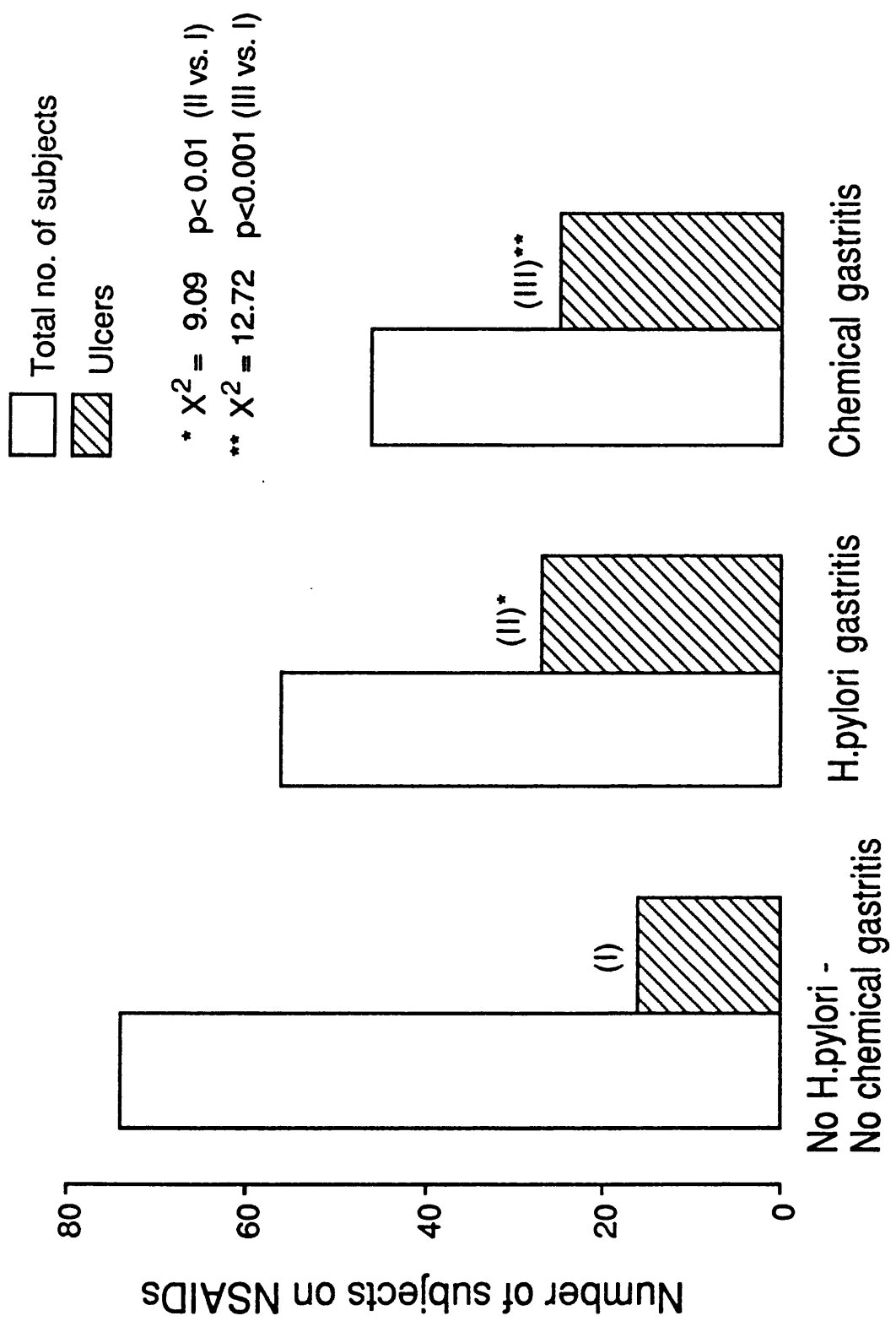


FIGURE 8

The distribution of NSAID related ulcers in patients with chemical gastritis, H pylori positive gastritis, and in those with neither of these conditions.



7.5 DISCUSSION

This study shows that NSAID patients have a higher prevalence of chemical gastritis, but lower prevalence of H pylori than subjects not on NSAID. The presence of chemical gastritis or H pylori seems to increase the likelihood of having NSAID related peptic ulceration. Also, there is a positive correlation between the endoscopic and the histological chemical scores in subjects taking NSAID.

Histological appearances, now considered as consistent with chemical gastritis, were previously reported in patients with bile reflux, following gastro-duodenal surgery (72,152). Subsequent studies have, on the other hand, found that there was no correlation between the reflux of bile and the development of chemical gastritis in the intact stomach (71). This would suggest that bile reflux has not significantly influenced the prevalence of chemical gastritis in this study. The author believes that he has corrected for the possible effect of bile by including in the analyses patients who were not taking NSAID.

Features of chemical gastritis were also described in patients with alcoholic haemorrhagic gastritis (153). In this study, it is interesting to find that 2 cases of chemical gastritis, in patients not on NSAID, had a history of heavy alcohol intake. The development of chemical gastritis in the NSAID group is unlikely to have been totally related to alcohol consumption, as judged from the demographic data of patients in this study.

The prevalence of chemical gastritis in this study is comparable

to that found by another group (71). The scores regarded as being attributable to chemical effect in this study were lower than those of Sobala et al. (71), this being justified on the basis of some possible differences in interpretation between the two centres. Moreover, despite being on the lower side, the chemical scores in this study still correlated positively with the endoscopic scores. Had the study included only those patients with high chemical scores (greater than 8 or 9), the prevalence of chemical gastritis would have been lower, but the correlation with the endoscopic scores would have become even more significant. It could not be accepted, however, that patients with chemical scores of 8 or 9 should be considered as having normal gastric histology.

In addition to chemical gastritis, NSAID have several other activities which can lead to gastric mucosal damage. These include inhibition of prostaglandin synthesis (7), interference with mucosal blood flow (154, Chapter 10), and disruption of the gastric mucosal barrier in a manner that allows back diffusion of hydrogen ion with its damaging consequences (11,12). Despite the considerable interest recently shown in some of the above mechanisms, probably because of their therapeutic potential (41), no correlation was found between peptic damage and activities of NSAID such as prostaglandin inhibition (5,8). This should emphasize the importance of identifying chemical gastritis, because of the positive correlation between the endoscopic and the histological chemical scores, as shown by this study. Chemical gastritis alone is unlikely to explain all cases of NSAID related peptic ulceration, probably due to the fact that multiple mechanisms are involved in such cases (7,11,12,154).

A link, probably causal, has been thought to exist between gastritis and gastric ulcers, as suggested by several previous studies (155-159). Such a relationship has been extended to include cases of gastritis found in association with *H pylori* (158-159). The situation in patients taking NSAID has not been clear: it might have been complicated by not taking into consideration the presence of chemical gastritis (160), owing to the failure of Whitehead's classification (122) to cover such histological entity. The finding of a greater number of NSAID related gastric ulcers in patients with chemical gastritis strengthens the broader relationship between gastritis in general and gastric ulcers (155-159). Patients on NSAID but without *H pylori* or chemical gastritis, had a total of 56 cases of gastritis (56/72, 78%), but the least number of ulcers. This might suggest that gastritis bears a strong relationship to NSAID-induced gastric ulcers only when it is chemical in nature, or if it is association with *H pylori*.

NSAID patients had a smaller proportion of cases positive for *H pylori* than those not on NSAID. The reason for this is not clear. It could be speculated that NSAID might have a direct toxic activity against *H pylori* or an indirect effect by altering the natural habitat of these organisms. NSAID were previously shown to affect the secretion rate and/or the characteristics of the mucus layer in the stomach. Aspirin (16) and indomethacin (17) were found to inhibit mucus secretion. It was also suggested that aspirin could increase pepsin-mediated proteolysis of mucus, decrease mucus viscosity and increase the permeability of mucus to hydrogen ion (18). Indomethacin was also found to inhibit active bicarbonate secretion by the gastric mucosa (19). The mucus layer, commonly accepted as

the natural habitat of *H pylori* (159), might be rendered less hospitable by the above changes.

In the NSAID group, patients positive for *H pylori* had a greater number of ulcers than those without *H pylori* or chemical gastritis. This might suggest a synergistic effect between NSAID and *H pylori* in causing gastric damage. Examples of this synergism were previously demonstrated with other bacteria in the intestine of laboratory animals. It was observed that the ulcerogenic effect of NSAID could be reduced by antibiotic treatment (20). Germ-free animals were also found to be resistant to indomethacin-induced intestinal lesions (21). More recently, the author has demonstrated that the combination of indomethacin and *H pylori* culture filtrate reduced human gastric mucosal viability and its PGE₂ production to a greater extent than indomethacin alone, in vitro (22). On the other hand, it is worth noting that, of 58 gastric ulcers identified in patients taking NSAID in this current study, only 20 (37%) were positive for *H pylori*. This is lower than the previously reported prevalence of *H pylori* in gastric ulceration unrelated to NSAID, which generally varied between 63% (161) and 77% (112). It is, however, consistent with other findings in this study, which suggest that NSAID might reduce the prevalence of *H pylori* infection. One cannot exclude the possibility that at least some of the ulcer patients in this study might have become negative for *H pylori* at some stage during their long term intake of NSAID, although there was no evidence of this in the study.

In conclusion, chemical gastritis occurs more commonly in patients receiving NSAID than in subjects who do not take these

agents. The importance of identifying this histological entity stems from its positive correlation with NSAID-related gastric ulceration. Although H pylori was isolated in only 32% of subjects on NSAID, its presence was associated with a greater number of ulcers than in NSAID patients without H pylori or chemical gastritis. These findings might contribute to our understanding of the interaction between NSAID and H pylori

CHAPTER 8

8. NSAID AND THE DUODENAL MUCOSA

8.1 SUMMARY

Background: Duodenitis and gastric metaplasia, which is often colonized by *Helicobacter pylori*, are increasingly recognized for their importance in the pathogenesis of duodenal ulcers. The situation is not clear in patients receiving non-steroidal anti-inflammatory drugs, who have a higher risk of peptic ulceration.

Aims:: To identify the duodenal histological abnormalities in the presence or absence of NSAID, *H pylori*, and duodenal ulceration.

Methods: Endoscopic duodenal biopsies were taken from healthy looking mucosa of 172 patients (74 took NSAID, and 98 did not). Duodenitis was graded according to the degree of neutrophilic and plasma cell infiltration, villus height, Brunner's gland prolapse, and gastric metaplasia. The activity of duodenitis was dependent on the neutrophilic infiltration. A global score covering all the above factors was constructed, and *H pylori* in both the stomach and duodenum, was assessed.

Results: Duodenitis with varying degrees of neutrophilic infiltration and gastric metaplasia was found in 20 patients (27%) taking NSAID, compared with 56 patients (57%) not on NSAID ($X^2 = 16.24$, $p < 0.001$). This degree of duodenitis was also found in 20 out of 25 patients (80%) with duodenal ulcers, regardless of NSAID intake ($X^2 = 15.38$, $p < 0.001$). Gastric metaplasia was identified in 20 patients (27%) on NSAID, and 38 (39%) not on NSAID.

Duodenal H pylori was only seen in patients with gastric metaplasia (10, 50% on NSAID and 34, 89% not on NSAID). H pylori-positive gastritis, and the combination of active duodenitis and gastric metaplasia were independent predictors of duodenal ulceration.

Conclusion: Active duodenitis is less common in patients taking NSAID, but is strongly associated with gastric metaplasia, H pylori positive gastritis and duodenal ulceration. These findings are relevant to the pathogenesis and the treatment of duodenal ulcers in patients taking NSAID.

8.2 INTRODUCTION

Gastritis and duodenitis have, for a long time, been thought to play a role in the pathogenesis of peptic ulcer disease (155-159, 162, 163) and such concept has been further consolidated by the recognition of Helicobacter (H) pylori (158-159). Subsequently, it has been suggested that gastric metaplasia in the duodenum and H pylori associated gastritis might be synergistic in the pathogenesis of duodenitis, with the metaplastic gastric epithelium allowing H pylori

to colonise the duodenal mucosa, where it produces an acute inflammatory response (164). The majority of studies in this field have been carried out in patients not taking NSAID and the role of duodenitis and gastric metaplasia in mediating NSAID - related damage has, therefore, remained unclear. This study aimed at studying the histology of the duodenal mucosa in the presence or absence of chronic NSAID intake, H pylori and duodenal ulcers.

8.3 PATIENTS AND METHODS:

Patients, aged 18 years or over, were recruited from the Rheumatology and Gastroenterology out-patient clinics, provided they had no previous history of gastric or duodenal surgery. NSAID had to be taken for a minimum of 4 weeks prior to endoscopy. Patients were excluded if they had taken ulcer-healing agents, antibiotics, or cytotoxic drugs within one week of endoscopy. Informed consent was obtained, and endoscopy performed using 3-7mg midazolam intravenously for sedation. At endoscopy, an average of two biopsies were taken from the anterior and posterior walls of the first part of the duodenum, and immediately fixed in a solution of 10% formal/saline. The duodenal histology was reported as follows: Normal, with villus to crypt ratio greater than 2:1, in the presence or absence of Brunner's glands above the muscularis mucosa, but without any increase in the numbers of inflammatory cells. Chronic duodenitis: in the presence of mucosal mononuclear cell infiltration and epithelial damage. Active chronic duodenitis: this was diagnosed whenever intramucosal neutrophils were seen, in the presence or absence of gastric metaplasia, and especially in the backbround of mononuclear cell infiltration. The severity of the above findings (duodenitis, villus height, and Brunner's gland prolapse)

was graded on a 0-3 scale: 0, normal; 1, mild; 2, moderate; and 3, severe. The extent of gastric metaplasia was assessed as follows: 0, absent; 1, present but consisting of less than 5 consecutive cells; 2, 5 consecutive cells; and 3, greater than 5 consecutive cells, seen in 1-3 sections cut at 3 levels in each biopsy (Figure 9).

A global score covering the overall totals of the scores of all the above assessments was constructed: in the absence of neutrophils, patients scoring 4 or less were considered normal; those scoring 5 or more were considered to have chronic duodenitis, in the absence of neutrophils. Chronic active duodenitis was diagnosed when intramucosal neutrophils were seen in patients scoring 4 or more.

The presence or absence of *H pylori* in the duodenal mucosa was also assessed using cresyl violet stain, and by culture of single duodenal biopsies. Detection of *H pylori* by either histology or culture was considered as indicative of active infection in the duodenum.

Gastric antral biopsies were also taken to check for the presence of *H pylori* by histology (2 specimens) and culture (1 specimen), as previously described (61). Gastritis was classified according to the Whitehead system modified to cover chemical and lymphocytic gastritis (23, Chapter 7).

Statistical analyses included the Chi-square (X^2) test, analysis of variance, multiple and logistic regression where appropriate. All specimens carried code numbers, and the endoscopist and pathologist were not aware of the patient's medications.

FIGURE 9

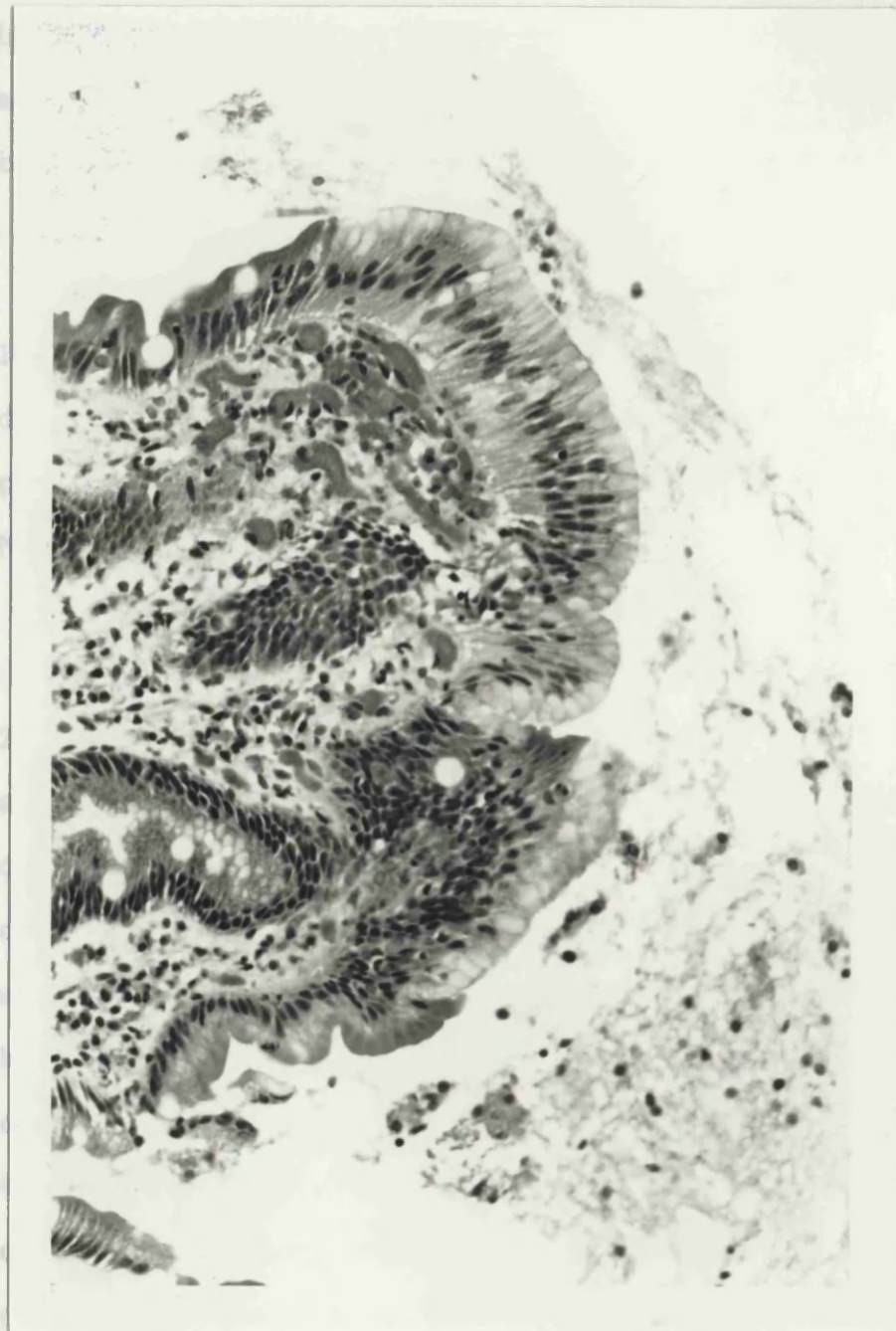
**A section through the duodenal mucosa, showing inflammatory
cell infiltration, and gastric metaplasia.**

Magnification X 110 (124a) ; X 274 (124b)



8.4 RESULTS

A total of 172 patients were studied. 74 were on NSAID, and 98 were not. Their demographic data are shown in Table 19. Patients



coexisted with gastric metaplasia.

The characteristics of patients with gastric metaplasia are shown in Table 21; although there were relatively fewer cases of metaplasia

8.4 RESULTS

A total of 172 patients were studied: 74 were on NSAID, and 98 were not. Their demographic data are shown in Table 19. Patients in both groups were comparable in their ages and in their smoking and drinking habits. Patients treated with NSAID had more gastric but similar number of duodenal ulcers to those not taking NSAID.

The duodenal global histological scores are illustrated in Figure 10. Patients in the NSAID group had fewer cases of active chronic duodenitis. There was no significant difference in the number of patients with chronic duodenitis (11 on NSAID and 10 not on NSAID).

Patients with active chronic duodenitis are described in Table 20. The two groups were similar with respect to the number of smokers, patients with gastric H pylori, gastric metaplasia, and duodenal ulcers. However, grade 3 (heavy) neutrophilic and plasma cell infiltration was more prevalent in the absence of NSAID. It is also worth noting that patients with active chronic duodenitis in both groups had a total of 20 out of 25 duodenal ulcer (80%) diagnosed in all patients. ($X^2=15.38$, $p < 0.001$, versus patients without active duodenitis). Duodenal H pylori was found in only 50% of NSAID patients with active duodenitis, and 61% of their counterparts not taking NSAID; all cases of duodenal H pylori coexisted with gastric metaplasia.

The characteristics of patients with gastric metaplasia are shown in Table 21; although there were relatively fewer cases of metaplasia

in patients taking NSAID, the differences were not statistically significant. Also, patients not treated with NSAID were more likely to have heavy neutrophilic and mononuclear cell infiltration. However, the two groups were almost identical in the extent of metaplasia, the number of smokers, patients with gastric H pylori and duodenal ulcers. Not unlike patients with active chronic duodenitis, the majority of duodenal ulcers (16/25, 64%) were also found to have gastric metaplasia ($X^2 = 14.00$, $p < 0.001$).

The prevalence of active duodenitis and gastric metaplasia in the presence or absence of the various types of gastritis is demonstrated in Table 22. The majority of cases were found in association with H pylori-positive gastritis, regardless of NSAID intake, although H pylori was less common in patients receiving NSAID. Active duodenitis was uncommon in patients with normal gastric histology: its prevalence varied between 20% (on NSAID) and 24% (not on NSAID). It is also worth noting that 24 of 25 duodenal ulcers (96%), found in all patients, were associated with active antral gastritis, and that gastric H pylori was identified in 23 of duodenal ulcer patients (92%). Logistic regression showed that gastric H pylori and the combination of active duodenitis and gastric metaplasia were independent predictors of duodenal ulceration. On the other hand, only 11 (9 on NSAID) of 16 gastric ulcers (69%) had H pylori related gastritis; the remaining 5 gastric ulcers were found in association with chemical gastritis, which was found only in patients taking NSAID.

The distribution of NSAID, with or without second-line drugs in patients with active duodenitis and gastric metaplasia is shown in Table 23. The median duration of NSAID intake was 3 years, and second-line drugs 2 years. Indomethacin, naproxen and diclofenac were the most widely used NSAID. Patients taking diclofenac, ketoprofen or nabumetone were less likely to have active duodenitis and gastric metaplasia than other NSAID but the differences were not statistically significant. Also, the intake of second-line drugs did not seem to influence the development of active duodenitis and gastric metaplasia, although such histological abnormalities, as well as gastric H pylori, were less common in patients treated with gold injections.

FIGURE 10

The duodenal global histological scores in the presence or absence of NSAID. Patients with intramucosal neutrophils have active chronic duodenitis. The remainder (black dots) have inactive chronic inflammation, and their numbers are similar in the two study groups.

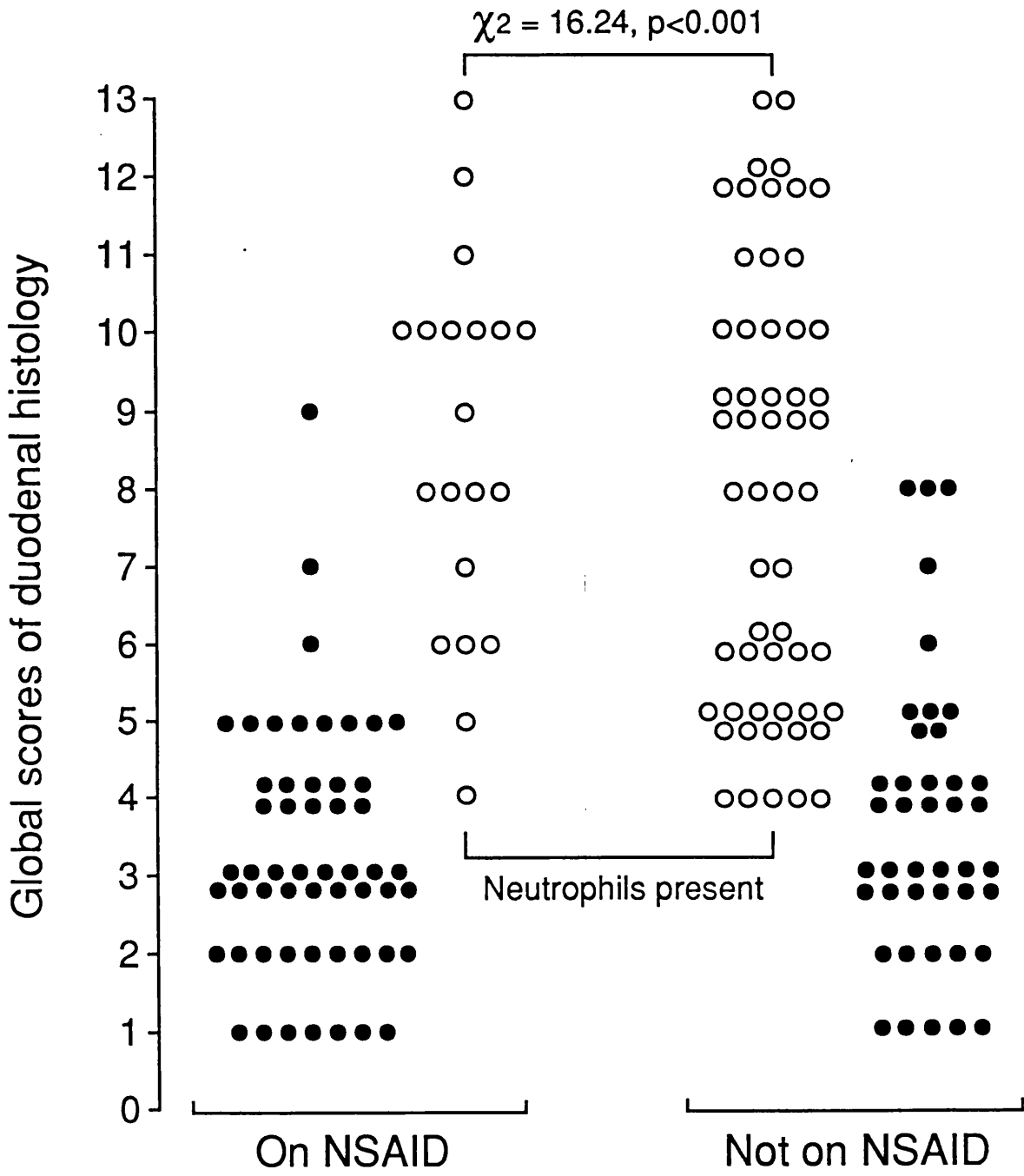


TABLE 19

DEMOGRAPHIC AND ENDOSCOPIC DETAILS OF THE STUDY GROUPS

	<u>ON NSAID</u>	<u>NOT ON NSAID</u>
Number	74	98
Males	24	38
Females	50	60
Age (years), median	57	52
(interquartile range)	(48-65)	(40-61)
Smokers	27	56
Drinkers	28	44
Abdominal symptoms	43	83
Gastric ulcers	17	2
Duodenal ulcers	11	14
Gastric erosions	7	-

TABLE 20

CHARACTERISTICS OF PATIENTS DUODENITIS No. (%)	WITH	ACTIVE	CHRONIC
	ON NSAID (N=74)	NOT ON NSAID (N=98)	
Number	20 (27%)	56 (57%)*	
Smokers	12 (60%)	37 (66%)	
Gastric H pylori	20 (100%)	43 (77%)	
Duodenal H pylori	10 (50%)	34 (61%)	
Gastric metaplasia	14 (70%)	34 (61%)	
Heavy neutrophilic infiltration	4 (20%)	17 (30%)**	
Heavy mononuclear infiltration	2 (10%)	12 (21%)**	
Duodenal ulcers	6 (30%)	14 (25%)	

SIGNIFICANT RISE: * : p <0.001 ; ** : p <0.05

TABLE 21: DETAILS OF PATIENTS WITH GASTRIC METAPLASIA IN THE

	DUODENUM	
	No. (%)	
	ON NSAID (N=74)	NOT ON NSAID (N=98)
Number	20 (27%)	38 (39%)
Grade: 1	4 (20%)	6 (16%)
3	16 (80%)	32 (84%)
Smokers	12 (60%)	24 (62%)
Gastric H pylori	14 (70%)	34 (89%)
Duodenal H pylori	10 (50%)	34 (89%)
Active chronic duodenitis	14 (70%)	34 (89%)
Heavy neutrophilic infiltration	4 (20%)	15 (39%)
Heavy mononuclear infiltration	1 (5%)	8 (24%)
Duodenal ulcers	6 (30%)	10 (26%)

TABLE 22: ACTIVE CHRONIC DUODENITIS AND GASTRIC METAPLASIA IN PATIENTS WITH OR WITHOUT GASTRITIS

	On NSAID (n=74)			Not on NSAID (n=98)		
	No.	Gastric H pylori duodenitis metaplasia	Active Gastric	No.	Gastric H pylori duodenitis metaplasia	Active Gastric
Chronic superficial gastritis	48	31	9	63	54	46
Chemical gastritis	10	-	4	-	-	-
Lymphocytic gastritis	3	3	3	7	5	3
Chronic atrophic gastritis	3	-	2	7	1	2
Normal	10	-	2	21	2	5

TABLE 23

DETAILS OF NSAID AND SECOND-LINE DRUGS IN PATIENTS WITH
ACTIVE CHRONIC DUODENITIS AND GASTRIC METAPLASIA

	No.	Gastric H pylori	Active duodenitis	Gastric metaplasia
Indomethacin	13	6	5	4
Naproxen	12	6	5	5
Diclofenac	7	2	-	-
Ketoprofen	6	3	-	1
Nabumetone	6	5	2	1
Ibuprofen	5	2	1	2
Others*	24	10	5	6
Sulphasalazine	18	14	12	8
Gold (im)	13	5	5	2
Penicillamine	6	2	3	1

*: these included 4 patients or less per each of the following agents:

azapropazone, fenbufen, flurbiprofen, piroxicam, etodolac, tiaprofenic acid, and sulindac

8.5 DISCUSSION:

This study shows that active chronic duodenitis is less common in patients treated with NSAID, and that the prevalence of gastric metaplasia in the duodenum is not significantly reduced by chronic NSAID use. The study also demonstrates a strong association between duodenal ulcers, active duodenitis, gastric metaplasia, and H pylori related gastritis regardless of NSAID intake.

Lesions identified as active chronic duodenitis in patients not taking NSAID were more frequently diagnosed in this study than that reported by some workers (164), who studied this abnormality in patients with non-ulcer dyspepsia. This could be explained by the presence of fewer ulcers in their study, the intercentre variation, and the nature of the scoring system of the current study. Such differences should not, however, influence the author's interpretation of the findings and his subsequent conclusions with respect to the effect of NSAID on duodenal histology, because of two main reasons. Firstly, the same global score was applied to biopsies taken from all patients, regardless of NSAID intake, under blinded conditions. Secondly, and allowing for the possibility that the global score had detected minor increases in the mucosal neutrophils (mild duodenitis) in some cases, the number of patients with heavy neutrophilic infiltration (severe duodenitis) was still greater in the absence of NSAID.

Gastric metaplasia tended to occur less frequently in NSAID patients, but the differences did not reach statistical significance.

This together with its close association with active duodenitis is in agreement with the findings of two recent studies (165,166). High acidity of the duodenal contents has for a long time been found to be associated with gastric metaplasia, both in man (164,165,167,168) and in the laboratory animals (159). NSAID are also known for their capacity to stimulate gastric acid secretion (13-15,26), which, at least in theory, should increase the prevalence of gastric metaplasia. The latter has not been observed in this study or in others (165,166) and could be related to the possible interaction between NSAID and H pylori. It has recently been proposed that the inflammatory injury to the duodenal mucosa by H pylori may stimulate the development of further gastric metaplasia (166). However, the prevalence of H pylori might be lower in chronic NSAID users, and this in turn might explain the lower than expected prevalence of gastric metaplasia in the presence of NSAID. Alternatively, gastric acid might not be elevated when NSAID are taken on long term basis, 3 years in this study.

Active chronic duodenitis was also less common in patients taking NSAID. The reason for this is not clear but it could be explained by the low prevalence of H pylori in NSAID patients, discussed above, and by the tendency of NSAID related damage to be maximal in the gastric antrum (42,82, Chapter 7). Although no specific histological picture could be demonstrated in the duodenal mucosa of NSAID patients, the relative lack of heavy neutrophilic and plasma cell infiltration in the duodenum is reminiscent of some aspects of chemical gastritis (23,71, Chapter 7). In the absence of gastroduodenal surgery, the latter is mostly found in chronic NSAID users, as shown in this study and others (23,71, Chapter 7).

Patients with active duodenitis had comparable proportions of cases with gastric *H pylori*, regardless of NSAID intake, and despite the differences in the overall prevalence of *H pylori* in the presence or absence of NSAID. This emphasizes the role of *H pylori* in the pathogenesis of active duodenitis (166). It is worth noting that in the presence of gastric metaplasia *H pylori* was almost as reliably isolated from the duodenum as from the stomach and this reflects the natural history of this infection, being dependent on the presence of gastric type epithelium. Since gastric metaplasia was found in the duodenum of only a limited number of patients, duodenal *H pylori* could not be taken as the only evidence of active infection with these organisms.

Patients receiving NSAID had 11 duodenal ulcers (11/74, 15%) compared with 14/98 (14%) in those not on NSAID. Similar numbers of duodenal ulcers (25-30%) were also seen in patients with active duodenitis and/or gastric metaplasia, regardless of NSAID intake. However, the majority of patients with duodenal ulcers, in the presence or absence of NSAID, were found to have active duodenitis (80%) and gastric metaplasia (64%). The strong association between such histological and endoscopic entities is in agreement with the suggestion that duodenitis and duodenal ulceration might represent different points in a disease spectrum with a common underlying pathogenesis (162-166). There was also a strong association between active duodenitis and *H pylori* positive gastritis, which in turn highlights the potential benefits of eradicating *H pylori* in minimising duodenal damage in NSAID patients. The same would apply to gastric ulcers, 8 of which (8/14, 57%) were positive for *H pylori* (23).

The relative rarity of active duodenitis in the NSAID group might explain, at least in part, why duodenal ulcers occur less commonly than gastric ulcers in such patients (42,82). On the other hand, duodenal ulcers were still observed in comparable proportions in the presence or absence of NSAID, which could be due to other mechanisms of NSAID toxicity such as suppression of mucosal prostaglandins.

The prevalence of gastric H pylori, active duodenitis and gastric metaplasia was similar in patients taking the various NSAID with the exception of diclofenac and ketoprofen. The prevalence of such findings tended to be lower in patients treated with gold injections than in those receiving sulphasalazine and NSAID (Chapter 12). A firm statement could not, however, be made because of the relatively small numbers of patients taking the individual NSAID with or without second-line drugs in this study.

In conclusion, despite the relative lack of active duodenitis in NSAID patients, a strong association exists between duodenal ulcers, active duodenitis, gastric metaplasia and H pylori positive gastritis regardless of NSAID intake. This might be relevant to the understanding of the pathogenesis and the treatment of duodenal ulcers in chronic NSAID users.

CHAPTER 9

9 NSAID AND THE OESOPHAGUS

9.1 SUMMARY

Background: Despite their strong association with gastric and duodenal ulcers, the effect of non-steroidal anti-inflammatory drugs (NSAID) on the oesophagus remains unclear. Conflicting results have been obtained from studies conducted on laboratory animals or small numbers of human volunteers.

Aims: To identify the endoscopic and histological abnormalities of the oesophageal mucosa, in the presence or absence of chronic NSAID therapy.

Patients and Methods: Seven-hundred and fifty-two patients were endoscoped; 596 took NSAID and 156 did not. All patients not in the NSAID group had abdominal complaints compared with 187 on NSAID. Histological studies were performed on normal looking mucosa of 98 patients (53 on NSAID and 45 not on NSAID).

Results: The overall prevalence of endoscopic oesophagitis in NSAID patients was 46/596 (8%). However, in symptomatic patients, endoscopic oesophagitis was found in 46/187 (25%) taking NSAID, compared with 19/156 (12%) not on NSAID ($X^2=9.8$, $p<0.001$).

At histology, four patients(7%) in the NSAID group (n=53) had papillary elongation and two (4%) had basal cell hyperplasia, compared with 13 (29%) and eight (18%) respectively, in patients not taking NSAID (n=45).

Conclusions: Endoscopic oesophagitis is more commonly found in chronic NSAID users with abdominal complaints. However, the relative lack of papillary elongation and basal cell hyperplasia might suggest that histological oesophagitis is not essential for the development of NSAID-related ulcers or erosions. Alternative mechanisms of NSAID toxicity should also be considered.

9.2 INTRODUCTION

The oesophageal mucosa can be adversely affected by a variety of agents such as gastric acid, pepsin, and bile salts (170-179). Oesophageal injury has also been ascribed to many of the commonly used medications, ingested in therapeutic doses (180-183). Non-steroidal anti-inflammatory drugs (NSAID) are amongst the most commonly prescribed agents (94,95), and their use has been associated with gastric and duodenal ulcers in about 30% of patients (42,82,128). However, the effect of NSAID on the oesophageal mucosa is not clear. There have been some reports, mostly of individual cases, of oesophagitis and stricture formation in patients taking NSAID (180-188), but some authors believe that the oesophagus is relatively unaffected by traditional NSAID therapy (189), and the experimental data of others suggest that NSAID might even be protective to the oesophagus in certain circumstances such

as irradiation and acid-induced oesophagitis (190-195).

The aim of this study was to identify the endoscopic and histological abnormalities of the oesophageal mucosa in the presence or absence of long term treatment with NSAID.

9.3 PATIENTS AND METHODS:

Patients, aged 18 years or over, were recruited from the Rheumatology and Gastroenterology Units. NSAID had to be taken for at least four weeks before endoscopy. Second-line drugs, and immunosuppressants such as steroids, azathioprine, methotrexate etc., were allowed provided they were taken together with NSAID. Patients with various arthropathies were accepted, including rheumatoid arthritis, osteoarthritis, ankylosing spondylitis, polymyalgia rheumatica, and systemic lupus erythematosus. Abdominal complaints had to be present to justify endoscopy in patients not taking NSAID. Subjects were excluded if they had a history of upper gastrointestinal surgery, Crohn's disease, malignancy, irradiation, or recent anti-ulcer therapy.

Informed consent was obtained and patients were endoscoped after the intravenous administration of diazepam or midazolam for sedation. Endoscopic oesophagitis was divided into two main groups: mucosal erythema and oedema with or without erosions (Group I), and chronic conditions (Group II) including isolated deep ulceration, strictures, and Barrett's oesophagus. This modification of the Savary and Miller grading system (196) was intended to improve the reproducibility of measurements in a large cohort study, especially because of the difficulty in distinguishing between Grades II and III

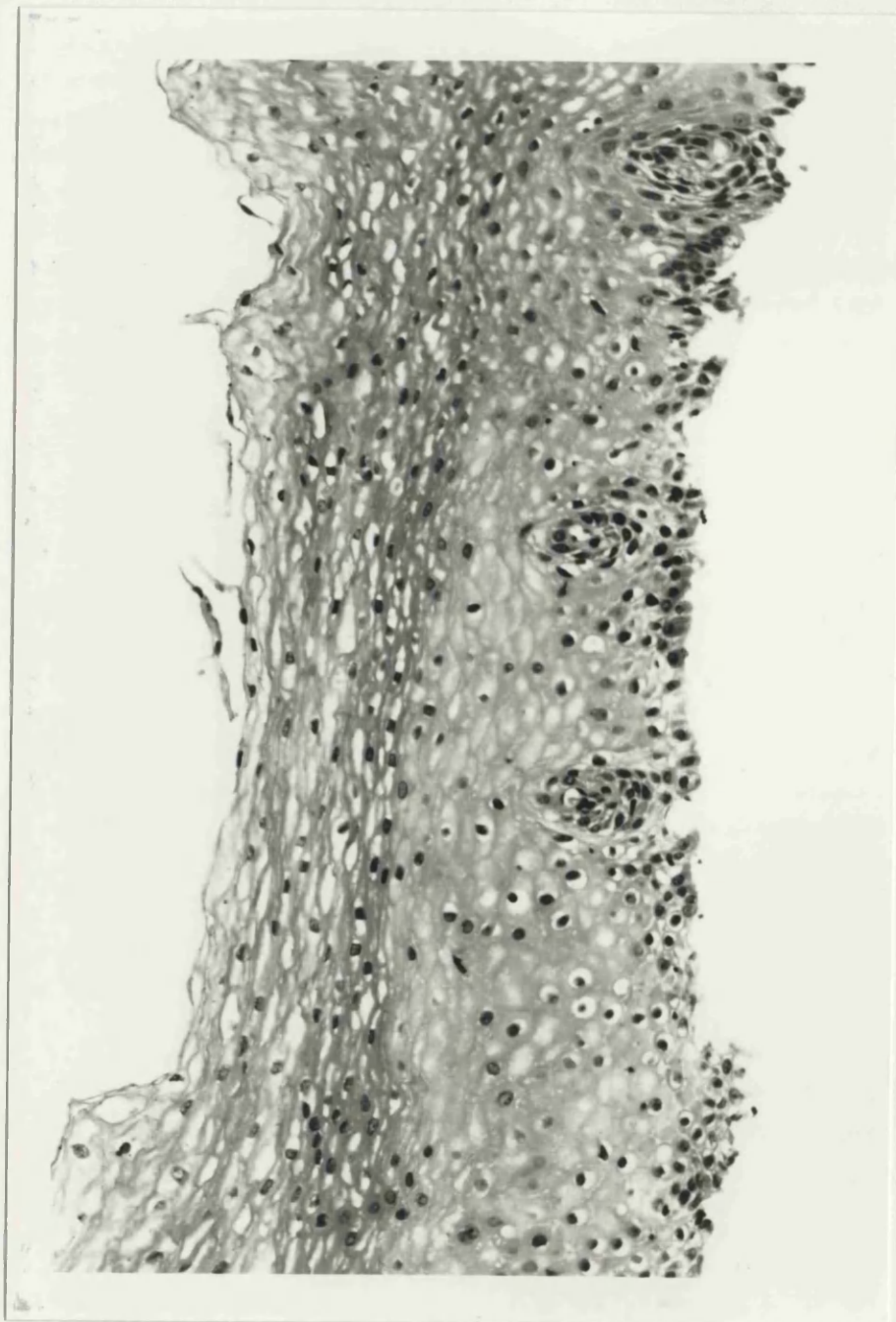
of Savary and Miller (196). The presence of other mucosal abnormalities of the stomach and duodenum was also recorded. Patients with the above oesophageal lesions and those with gastric ulcers had routine biopsies taken for histology. Special fungal stains were used whenever *Candida albicans* was suspected. Barrett's oesophagus was defined as gastric mucosa extending more than 3cm above the endoscopic gastro-oesophageal junction.

Histological study: The last 100 patients to be studied, who also had normal looking oesophagus, regardless of NSAID intake, had an average of two biopsies taken from the lower third of their oesophagus about 5cm from the cardia for histology. The following histological features of the oesophageal mucosa (Figures 11,12) were assessed: papillary elongation, basal cell hyperplasia, and inflammatory cell infiltration (197). The mucosal papillae were considered elongated if they measured greater than 60% of the distance between the basal cell layer and the luminal surface. Basal cell hyperplasia was defined as a condition in which the basal cells were greater than two layers in thickness. Inflammatory cell infiltration was considered heavy if at least five cells were seen per microscopic field of the whole biopsy specimen. Staining for *Candida albicans* was performed in all of the oesophageal biopsies. Gastric antral biopsies were also taken to check for *Helicobacter pylori* by both histology (two specimens) and culture (one specimen) as previously described (61).

Statistical analyses: These included the Chi-square (X^2) test and the analysis of variance, and the 95% confidence intervals for the estimated relative risk, where appropriate. The endoscopist, histologist, and bacteriologist were not aware of the details of patients' medications. The study was approved by the local Ethics Committee.

FIGURE 11

Normal oesophageal mucosa (X247)



(X300)

FIGURE 12

Some features of reflux oesophagitis: basal cell hyperplasia, and inflammatory cell infiltration.

The papillary size is at the upper limit of normal. (X395)

0.4 RESULTS:

Endoscopic study: A total of 282 patients underwent endoscopy: 596 took NSAID and 158 did not. These results are shown in Table 24. Both groups were comparable in age, smoking, and drinking habits. Because of the nature of the inclusion criteria



patients with oesophagitis, regarding abdominal pain, had upper abdominal complaints: dysphagia was present in a total of seven patients, four of whom were found to have ulcers. The rest of patients with oesophagitis complained of epigastric pain, heartburn and nausea with or without vomiting.

9.4 RESULTS:

Endoscopic study: A total of 752 patients underwent endoscopy: 596 took NSAID and 156 did not. Their details are shown in Table 24. Both groups were comparable in sex distribution, age, smoking, and drinking habits. Because of the nature of the inclusion criteria abdominal complaints were less prevalent in patients taking NSAID, but this group still had more ulcers and erosions affecting the stomach and duodenum. In the NSAID group, patients with or without abdominal complaints had similar demographic details, but gastric and duodenal lesions were commoner in the symptomatic group. The numbers of patients with oesophagitis and the details of the oesophageal lesions are illustrated in Figure 13. The overall prevalence of endoscopic oesophagitis was 46/596 (8%, NSAID present) and 19/156 (12%, NSAID absent). However, when abdominal complaints were taken into consideration, since all non-NSAID patients had to have such complaints to justify endoscopy, their symptomatic counterparts in the NSAID group (acting as case controls) had a higher prevalence of oesophagitis (46/187, 25%, $p < 0.001$). Intense erythema without erosions was found in 12 NSAID patients and 5 others not on NSAID. There were no significant differences in the prevalence of strictures or Barrett's oesophagus, but oesophagitis with or without erosions or deep ulcers was commoner in NSAID patients with abdominal complaints. All oesophageal lesions described in this study affected the lower third of oesophagus. Other characteristics of patients with oesophagitis are shown in Table 25. It is worth noting that all patients with oesophagitis, regardless of NSAID intake, had upper abdominal complaints: dysphagia was present in a total of seven patients, four of whom were found to have strictures. The rest of patients with oesophagitis complained of epigastric pain, heartburn and nausea with or without vomiting.

Rheumatoid arthritis was the commonest diagnosis amongst various patients taking NSAID (Table 26). Although oesophagitis was found in four of 27 patients (15%) with systemic lupus erythematosus, and 35/499 (7%) with rheumatoid arthritis, the differences between the two groups did not reach statistical significance.

Drugs taken by patients in the NSAID study groups are detailed in Tables 27 and 28. Indomethacin, naproxen, and diclofenac were the commonest NSAID used. The median duration of NSAID therapy was three years in all groups. Also, patients treated with prednisolone had a similar prevalence of oesophagitis to those treated with gold injections, sulphasalazine, or penicillamine. However, oesophagitis was found in 22 out of 206 patients (11%) treated with second-line drugs or immunosuppressants plus NSAID, compared with 24/390, 6% of those taking NSAID only ($\chi^2 = 5.4$, $p < 0.02$; ER:1.8; 95% CI:1-3.3). Oesophageal lesions in gold treated patients consisted of erythema (5 cases) and erosions (3 cases) but no ulcers; erosions were also found in all patients with oesophagitis who received other disease modifying drugs. Oesophagitis was not seen in any of the patients receiving hydroxychloroquine: such patients had a median age of 34 years, compared with 58 years in patients treated with other agents, and took NSAID for a median period of one year.

Histological study: One hundred patients with endoscopically normal looking oesophagus were studied; two were excluded because of recent use of H₂-receptor antagonists. Of the remaining 98, 53 patients took NSAID and 45 did not. Their characteristics are shown in Table 29. There were no significant differences between the two groups with respect to age, smoking and drinking habits. However, patients not taking NSAID had fewer gastric ulcers but a greater number of patients with duodenal ulcers and/or *Helicobacter*

pylori.

The prevalence of the main histological abnormalities, in the presence or absence of NSAID, is illustrated in Figure 14. Papillary elongation and basal cell hyperplasia were less common in patients treated with NSAID but the differences were not highly significant as judged by the confidence intervals. Also, the extent and degree of inflammatory cell infiltration was not influenced by NSAID intake. All 13 patients with papillary elongation not taking NSAID, and three out of four patients with this abnormality in the NSAID group, were heavy smokers. Heavy smoking was also present in six out of eight patients with basal cell hyperplasia not treated with NSAID, and one of two of their counterparts in the NSAID group.

TABLE 24

Details of all Patients in the Endoscopic Study. NSIAD patients are classified according to the presence (group I) or absence (group II) of abdominal complaints

	<u>On NSAID</u>			<u>Not on NSAID</u>
	<u>I</u>	<u>II</u>	<u>Total</u>	
Number	187	409	596	156
Male	62	96	158	61
Females	125	313	438	95
Age, Median	56	58	57	51
(interquartile range)	(43-62)	(49-67)	(43-65)	(47-62)
Smokers	88	142	230	70
Drinkers	68	92	160	71
Indication for endoscopy:				
Abdominal complaints	187	-	187	156
Anaemia	46	85	131	-
Weight loss	9	12	21	-
Gastric ulcers	37	46	83	6
Duodenal ulcers	33	21	54	16
Gastric/duodenal erosions	40	55	95	4

TABLE 25

Characteristics of Patients with Oesophagitis

	On NSAID	Not on NSAID
Number	46	19
Males	15	6
Females	31	13
Age, median	56	52
(interquartile range)	(47-65)	(36-72)
Smokers	10	7
Drinkers	9	9
Abdominal symptoms	46	19
Co-existing peptic ulcers		
Gastric	4	1
Duodenal	8	2
Oesophageal Candidiasis	3	2

TABLE 26

The Basic Diagnosis in Patients Taking NSAID

Diagnosis	All NSAID patients (n=596)	Patients with oesophagitis (n=46)	Patients in the histology study (n=53)
Rheumatoid			
arthritis	499	35	44
Osteoarthritis	29	1	9
Systemic lupus			
erythematosus	27	4	-
Ankylosing			
spondylitis	17	2	-
Polymyalgia			
rheumatica	8	-	-
Psoriatic			
arthropathy	7	1	-
Reactive arthritis	4	1	-
Gout	4	1	-
Henoch-Schonlein	1	1	-
purpura			

TABLE 27
Types of NSAID Used

Drugs	All NSAID patients (n=596)	Patients with oesophagitis (n=46)	Patients in the histology study (n=53)
Indomethacin	146	11	10
Naproxen	120	12	8
Diclofenac	110	9	6
Flurbiprofen	70	1	2
Ketoprofen	50	1	5
Ibuprofen	36	8	2
Piroxicam	24	1	4
Others*	40	3	16

* 3-6 Patients per one of the following:

Fenpufen, etodolac, azapropazone, naubmetone, sulindac,
tiaprofenic acid, tenoxicam, and tolmetin

Table 28

Details of Second-Line and Immunosuppressive Drugs Taken by NSAID
Patients

Drugs	All NSAID patients (n=596)	Patients with oesophagitis (n=46)	Patients in the histology study (n=53)
Gold injections	67	8	11
Sulphasalazine	34	4	10
Hydroxychloroquine	27	-	4
Penicillamine	24	3	4
Prednisolone	33	3	-
Azathioprine	11	2	-
Methotrexate	3	-	-
Cyclophosphamide	4	1	-
Cyclosporin	3	1	-

Table 29

Characteristics of Patients in the Histological Study

	On NSAID	Not on NSAID
Number	53	45
Males	13	19
Females	40	26
Age, median	58	52
(interquartile ranges)	(49-65)	(45-63)
Smokers	26	31
Drinkers	16	18
Gastric ulcers	8	2
Duodenal ulcers	5	10
Gastric <i>Helicobacter pylori</i>	31	36

FIGURE 13

Endoscopic oesophageal lesions in patients with abdominal complaints, in the presence or absence of NSAID:

- * $p < 0.001$ (estimated relative risk, E.R., 2.35; 95% confidence intervals, C.I.: 1.31, 4.212)
- ** $p < 0.01$ (ER: 2.42; C.I.: 1.25, 4.67) Intense erythema without erosions was found in 12 NSAID users and 5 non-users. This did not affect the overall levels of statistical significance
- *** $p < 0.02$ (ER, C.I., unmeasurable).

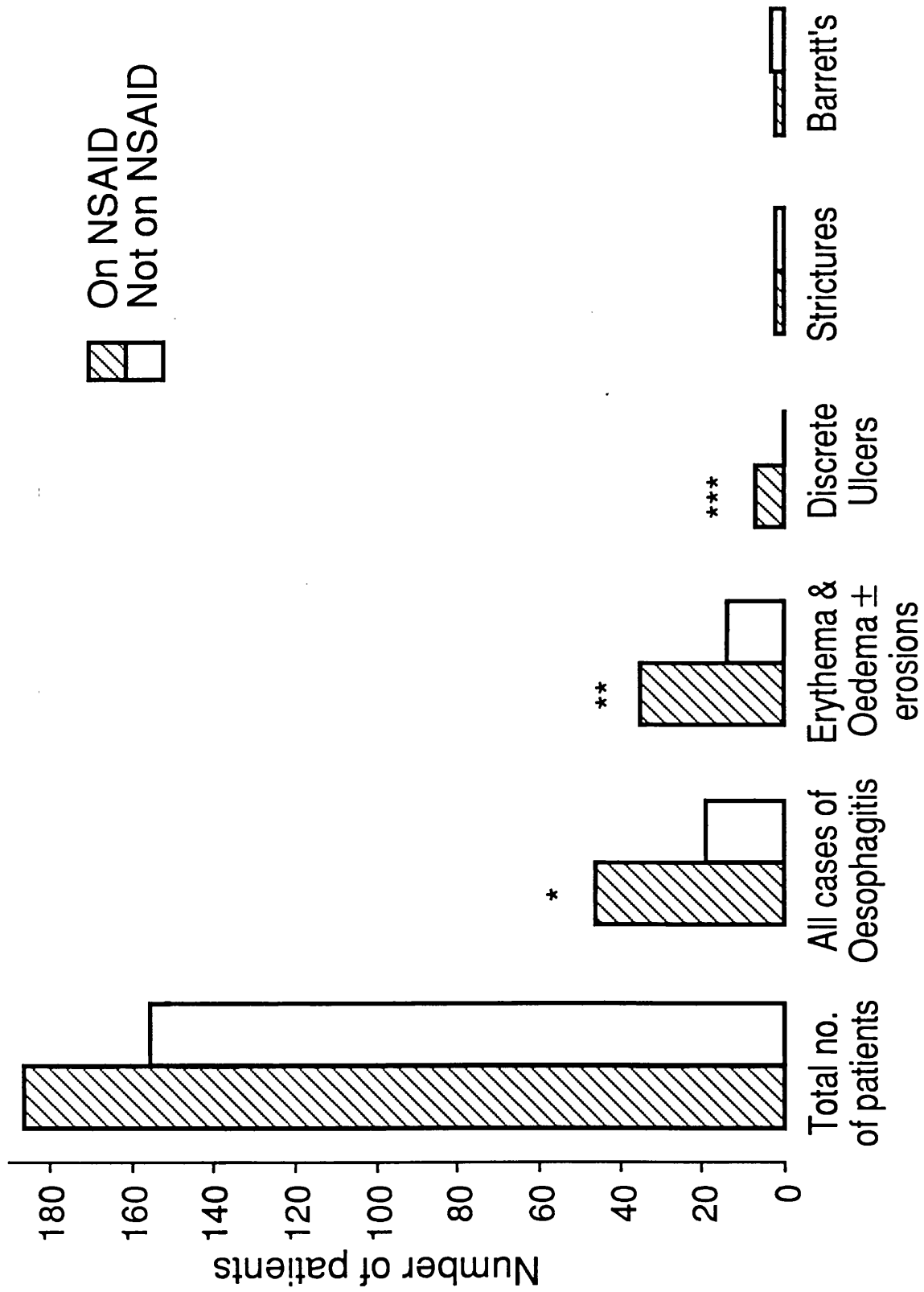
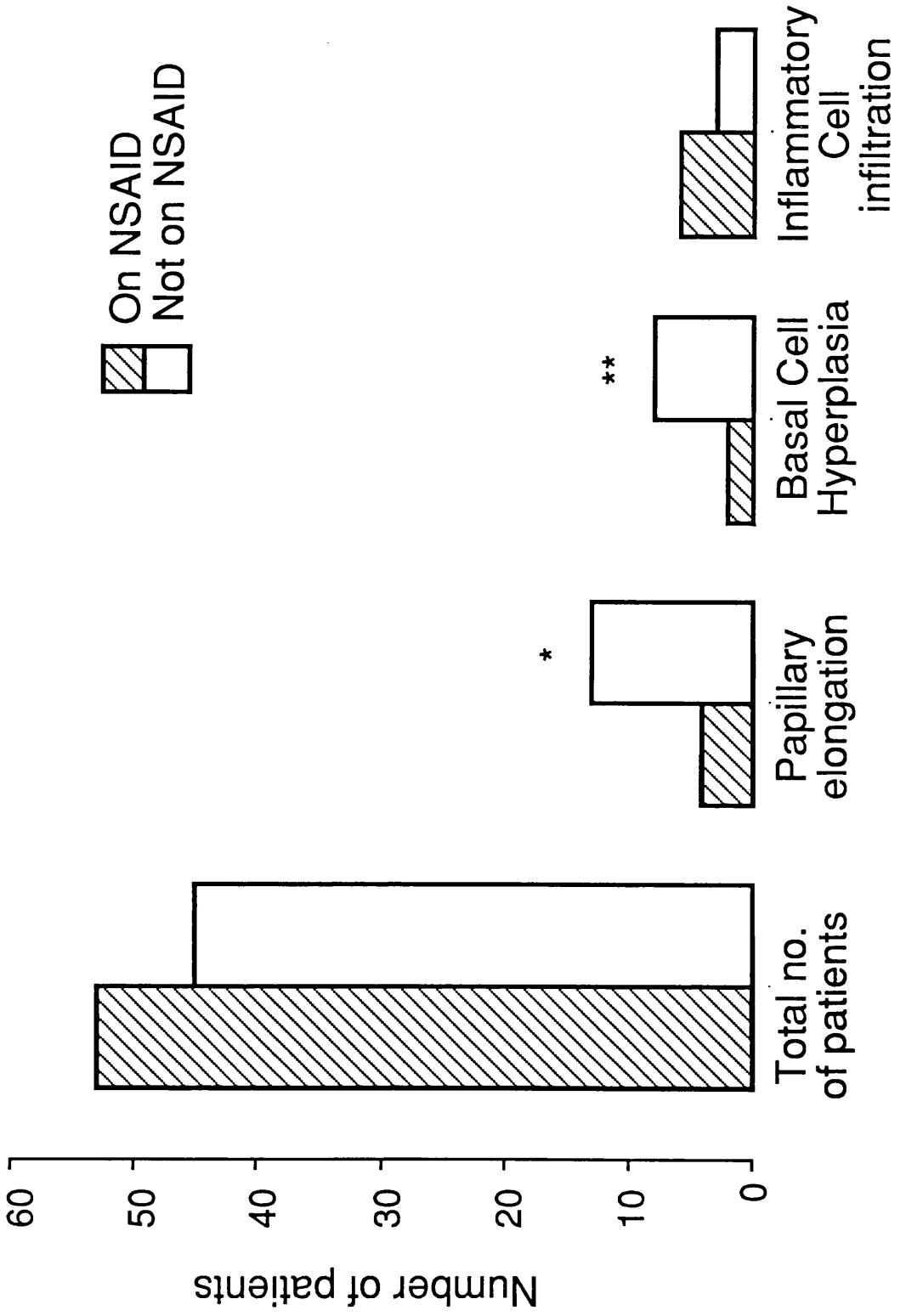


FIGURE 14

Oesophageal histological findings in the presence or absence of NSAID:

- * $p < 0.01$ (ER:0.201;C.I. :0.06,0.671)
- ** $p < 0.02$ (ER:0.18;C.I.: 0.036,0.897)

The p-values should be interpreted with caution because of the wide range of the 95% confidence intervals and the similarity in the estimated relative risk. The overall view is that the two study groups are comparable in their histological findings.



9.5 DISCUSSION:

This study shows that the prevalence of endoscopic oesophagitis, with or without ulcers or erosions, is higher in NSAID patients with upper abdominal complaints than in those not treated with such agents. The prevalence of strictures or Barrett's oesophagus is similar in both groups. Histological studies in patients with normal looking oesophagus at endoscopy, show that papillary elongation and basal cell hyperplasia are relatively less common in the presence of NSAID, although the extent and degree of inflammatory cell infiltration are similar in the presence or absence of NSAID intake.

Endoscopic oesophagitis is now a common disease and accounts for as many as 19% of new referrals for endoscopy (198). This is almost identical to the mean prevalence of oesophagitis (18.5%) in the author's symptomatic patients, regardless of NSAID intake (25%, NSAID present; 12%, NSAID absent; mean 18.5%). The overall prevalence of oesophagitis in NSAID patients was only 8%. The presence of abdominal complaints had to be taken into consideration because all patients not taking NSAID were symptomatic and acted as case controls. Ideally, asymptomatic subjects not treated with NSAID should have also been included, but ethical approval was not available.

In the few published studies on NSAID-related oesophagitis the prevalence of this endoscopic abnormality had varied between 15.7% (187) to 20% (186) which is comparable to that found in symptomatic NSAID patients in this study. Also, two out of four patients with oesophageal strictures, in this study, had taken NSAID compared with 31% found by another group (199). These findings do not support the idea that benign strictures are commoner in patients

treated with NSAID, although a firm statement cannot be made because of the small number of strictures involved.

Upper abdominal complaints were present in all patients with oesophagitis. These could be due to the oesophageal lesion itself, the co-existing peptic ulceration, or both. However, symptoms are not a consistent feature of NSAID-related gastric or duodenal ulcers, as found by this study and others (81,82).

Oesophagitis affected the lower third of oesophagus of all NSAID patients, confirming the findings of other studies in this field (137, 188). This is unlike many cases of oesophagitis related to other types of drugs, which affect the middle third of oesophagus (181,183). The reason for this is not clear, but it could be related to the method of diagnosis: barium studies might not be ideal for identifying erythema and small ulcers or erosions. It is interesting to find that oesophageal lesions were also seen in the lower third of oesophagus, as a result of medications not containing NSAID, when endoscopy was used (182). Endoscopy was, therefore, considered as the most reliable method of diagnosing drug related oesophagitis (180, 182, 200).

In patients with normal oesophagus at endoscopy, the histological assessments have shown that papillary elongation and/or basal cell hyperplasia are uncommon in the presence of a history of chronic NSAID intake. Such features are recognised as being the histological consequences of gastro-oesophageal reflux in man (197), and their relative rarity in the presence of NSAID raises two possibilities. Firstly, NSAID might have a beneficial effect in preventing or

minimising reflux oesophagitis, at least at the histological level. This has been suggested by the findings of studies conducted in cats, in which experimental oesophagitis was defined as polymorphonuclear infiltration and, in some cases, denudation of the epithelium (190). This is unlikely to be the case in this study, since the extent and degree of inflammatory infiltration were the same in the presence or absence of NSAID. Also, the duration of experimental oesophagitis might not have been long enough to allow for the evolution of hypertrophic and/or hyperplastic changes in the oesophageal mucosa.

Secondly, it could be speculated that histological oesophagitis is not essential for the development of NSAID-related ulcers or erosions. Other mechanisms of NSAID damage should therefore be considered, including their interaction with prostaglandins, and their direct toxic effects especially when tablets get lodged in the oesophagus. Prostaglandins are known for their hypertrophic activity in the gastrointestinal mucosa (201), although this has not been demonstrated in the oesophagus. The ability of NSAID to inhibit mucosal prostaglandins might account for the lower prevalence of papillary elongation and basal cell hyperplasia in patients receiving NSAID. Certain prostaglandins (E_1 , E_2 , and A) can also decrease the basal lower oesophageal sphincter pressure in man (202) and their inhibition by NSAID could theoretically reduce gastro-oesophageal reflux with its histological consequences. However, NSAID have other important properties that may contribute to their damaging effects on the oesophageal mucosa. These agents are relatively acidic molecules, and are lipid soluble at low pH. When the pH of the

lower oesophagus is below 4, at the time of gastro-oesophageal reflux, NSAID may enter the oesophageal mucosal cells, causing direct toxic effects (183). Thirdly, the possibility of some sampling errors should not be ruled out given the occasional difficulties in interpreting oesophageal biopsies.

Patients not taking NSAID had a higher prevalence of H pylori, duodenal ulcers, papillary elongation, and basal cell hyperplasia of the oesophageal mucosa. While the associations between duodenal ulcers and H pylori or oesophagitis are well-recognised, a direct link between H pylori and endoscopic or histological oesophagitis has not been proven.

The histological study also showed a strong association between smoking and both papillary elongation and basal cell hyperplasia. This could be explained by the recognised inhibitory effect of smoking on the lower oesophageal sphincter, which as a result, increases gastro-oesophageal reflux (203).

The possible effect of anti-rheumatoid second-line drugs on the oesophagus has not been studied before. Patients taking these agents or immunosuppressive therapy plus NSAID had a higher prevalence of oesophagitis than those treated with NSAID only, in this study. Although a direct toxic effect cannot be entirely ruled out, patients requiring second-line drugs or immunosuppressants tend to be old, and to have more aggressive underlying disease. Older subjects are at greater risk of drug-induced oesophageal injury because of the disordered contraction of the lower oesophageal sphincter (204). This might explain the absence of oesophagitis in rheumatoid patients receiving hydroxychloroquine as they were younger than those treated with other second-line drugs. In

addition, the number of cases of Candidal oesophagitis, known to affect immunocompromised subjects (181), was not high in patients receiving immunosuppressants in this study, probably because of the small number of patients who took such agents. The inclusion in this study of a variety of arthritic diseases is also unlikely to have affected the findings, as none of such diseases has been shown to affect the oesophagus.

In conclusion, endoscopic oesophagitis with ulcers or erosions is commoner in NSAID patients with abdominal complaints. The relative rarity of papillary elongation and basal cell hyperplasia in NSAID patients might suggest that ulcers and erosions are not dependent on histological oesophagitis for their development, and highlights the importance of alternative mechanisms such as direct mucosal toxicity.

SECTION V
NSAID AND MUCOSAL BLOOD FLOW

CHAPTER 10

10 GASTRIC AND DUODENAL MUCOSAL BLOOD FLOW IN PATIENTS RECEIVING NSAID; INFLUENCE OF AGE, SMOKING, ULCERATION, AND H PYLORI

10.1 SUMMARY

Background: Studies conducted in experimental animals have suggested that gastric mucosal blood flow is reduced by acute exposure to NSAID. The possible interaction with other factors, relevant to the pathogenesis of peptic ulcers, has not been studied.

Aims: To assess the possible influence, on mucosal blood flow, of chronic NSAID intake, age, smoking, ulceration, and H pylori.

Methods: Using laser Doppler flowmetry, gastric and duodenal mucosal blood flow was measured in 70 patients who had taken NSAID for longer than 4 weeks, and studied the correlation with demographic factors, ulceration, and Helicobacter pylori. Blood flow was also measured in 17 other subjects not taking any drugs. Measurements were taken from healthy looking mucosa in the gastric antrum and first part of duodenum.

Results: Both gastric and duodenal blood flow values were significantly lower in patients taking NSAID than in those who did not. In the NSAID group the median duodenal mucosal blood flow was 150 perfusion units in smokers (n=29) compared with 175 in non-smokers (p=0.024), 123 units in patients with duodenal ulcers

(n=12) compared with 160 in those without duodenal ulcers (p=0.020), 135 units in patients with H pylori (n=30) compared with 168 in patients without H pylori (p=0.033), and 118 in smokers infected with H pylori compared with 175 units in non-smokers not infected with H pylori (F=13.4, p=0.0005). There was no correlation with age. Gastric blood flow was not significantly influenced by any of the above variables.

Conclusion: These results suggest that chronic NSAID intake is associated with reduced blood flow in both the stomach and duodenum. However, amongst NSAID patients duodenal, but not gastric, mucosal blood flow is reduced in smokers, and in those with duodenal ulcers and Helicobacter pylori. Multivariate analysis showed that only the simultaneous presence of smoking and H pylori had an independent suppressive effect, and when combined, lower blood flow values are observed, which might suggest a synergistic relationship between these two factors.

10.2 INTRODUCTION

Mucosal blood flow is believed to play an important role in maintaining the integrity of the gastric mucosa. Mucosal ischaemia has been found to be the main cause of gastric lesions in patients with thermal or head injury (205). Interference with blood flow by stress, vascular occlusion, or non-steroidal anti inflammatory drugs (NSAID) is also thought to cause gastric ulceration in animals (206). The situation is not clear in patients on long term NSAID who might have other factors that can, at least in theory, influence gastric mucosal blood supply. Also, very little is known about duodenal blood flow in such patients. The main aim of this study was to

measure gastric and duodenal mucosal blood flow in patients receiving NSAID, and to see if this could be influenced by age, smoking, ulceration, or (H) pylori. A small number of patients not taking NSAID were also included in order to study the overall effect of NSAID on the gastric and duodenal mucosal blood flow.

10.3 PATIENTS AND METHODS:

Patients with osteoarthritis or rheumatoid arthritis were included if they had taken NSAID for a minimum of 4 weeks. They were excluded if they had a history of previous gastric surgery or if they took anti-ulcer therapy, hypotensive agents, or anti-anginal drugs. They were also excluded if they had other systemic diseases such as diabetes mellitus. Similar criteria were also applied to another group of subjects, not on NSAID, who acted as controls, and were being investigated for upper abdominal complaints. Informed consent was obtained and endoscopy was performed after an overnight fast. Midazolam, 4-10 mg intravenously, was used for sedation.

Blood flow was measured with a laser Doppler flowmeter (210,211) (Periflix PF3, Perimed, Sweden) which was regularly calibrated using a standard suspension of latex particles to ensure constant sensitivity. The fibre-optic probe (Model PF309) was introduced through the biopsy channel of the endoscope, and its tip applied to healthy-looking mucosa in the gastric antrum, within 5cm of the pyloric canal, and the first part of the duodenum. The antrum was chosen because it is the commonest site of NSAID-related gastric damage (5,42). In patients with ulcers, the probe was applied at least 2cm away from the ulcer edge. The contact pressure between the probe and tissue was the minimum necessary to

maintain optical coupling. In pilot experiments it was found that varying the contact pressure within reasonable limits did not have any consistent effects on recorded flow values, presumably because of the considerable depth of penetration of laser light in gastrointestinal tissue (Johansson et al,212).

The flowmeter was operated using the 12 kHz bandwidth and 3 sec time constant settings, and the output, in arbitrary perfusion units, was recorded continuously for a minimum period of one minute at each location on a chart recorder. Flow values were calculated as the mean flowmeter reading over this period, excluding any artefacts associated with movement of the probe or peristalsis. The reproducibility of measurements was assessed in pilot studies in which recordings were repeated in the same region of the stomach after a period of 10-15 minutes, and the coefficient of variation was found to be 17%. All recordings were analysed without knowledge of the patients' clinical details or endoscopic findings.

The presence of *H pylori* was determined by histology and bacteriology, as previously described (61), in biopsies taken from the gastric antrum. The histologist and the bacteriologist were not aware of the patients' details or other results.

Statistical tests included the Mann-Whitney, Kruskal-Wallis, and multiple linear regression analysis with step-wise variable elimination, where appropriate.

10.4 RESULTS

A total of 87 patients were studied: 70 took NSAID and 17 did not. Their characteristics are shown in Table 30. Mucosal blood flow values in the stomach, 125(90-183) and duodenum, 160(123-190) units, median (interquartile ranges), in patients taking NSAID, were lower than their corresponding values, 200(110-350) and 265 (150-310), in patients not taking NSAID ($p=0.01$). In NSAID patients with osteoarthritis, the gastric mucosal blood flow was 125 (90-160) units, and the duodenal blood flow was 148 (110-175), compared with 125 (90-185) and 160 (125-200) units, respectively, in those with rheumatoid arthritis. Therefore, there were no significant differences between patients with osteoarthritis or rheumatoid arthritis with respect to blood flow. In addition, there was no correlation between mucosal blood flow and parameters of arthritic disease activity such as the erythrocyte sedimentation rate or C-reactive protein.

H pylori was found in 10 of 15 NSAID patients (66%) with gastric ulcers (all affecting the antrum) and 7 of 12 patients (58%) with duodenal ulcers. Four of 15 gastric ulcer patients (27%) were smokers compared with 8 of 12 patients (67%) with duodenal ulcers. Only 2 patients in the control group had ulcers, both of which were in the duodenum.

Gastric and duodenal blood flow of NSAID patients in various age groups are shown in Table 31. Patients older than 60 years tended to have lower blood flow than those younger than 40 years. However, this tendency did not reach statistical significance.

Smoking, ulceration, or H pylori did not seem to affect gastric blood flow of patients taking NSAID in this study (Table 32). On the other hand, the presence of these factors was associated with lower blood flow values in the duodenal mucosa (Table 35). In the NSAID group as a whole, mucosal blood flow was 125 (90-183) units in the stomach compared with 160 (123-195) units in the duodenum ($p=0.00017$). To investigate which factors were independently associated with reduced blood flow in the duodenum, multiple linear regression analysis with step-wise variable elimination was performed. The three factors, (duodenal ulceration, smoking and H pylori) were assigned numerical values of 0 or 1 according to their presence or absence, and the predictor variables included in the analysis were the factors themselves and all two-and three-factor products. A square root transformation was performed on the duodenal blood flow values to correct an upward skewing of the distribution. The only variable with a statistically significant relationship with duodenal blood flow was the product (ie simultaneous presence) of smoking and H pylori: smokers infected with H pylori had a median (interquartile range) blood flow of 118 (85-168) compared with 175 (135-250) units in the rest of the NSAID group ($F=13.5$, $p=0.0005$). The independent effect of duodenal ulceration approached statistical significance ($F=3.3$, $p=0.076$).

In the small number of subjects not taking NSAID, no significant differences were found in response to H pylori, peptic ulceration, or the demographic details.

TABLE 30
 Characteristics of the Study Groups

	<u>On NSAID</u>	<u>Controls</u>
Total number	70	17
Males	23	6
Females	47	11
Median Age (years)	54	53
Smokers	29	11
Osteoarthritis	14	-
Rheumatoid arthritis	56	-
Duration of arthritis median (years)	7	-
Gastric ulcers	15	-
Duodenal ulcers	12	2
H pylori	30	13

TABLE 31

Gastric and Duodenal Blood Flow (Perfusion Units),
 Median (Interquartile Ranges), in NSAID Patients Classified
 According to their age Groups (Years)

	<u>Less than 40</u>	<u>41-60</u>	<u>Greater than 60</u>
Sample size	14	46	10
Gastric blood flow	155 (115-200)	120 (90-175)	115 (83-153)
Duodenal blood flow	158 (135-185)	163 (110-235)	145 (110-175)

TABLE 35
 Gastric Blood Flow (Perfusion Units) in NSAID Group
 (Interquartile Ranges) in the Presence of
 Smoking, Ulcers or H pylori

	Present		Absent	
	Sample Size	Value	Sample Size	Value
Smoking	29	123 (85-185)	41	125 (100-190)
Gastric Ulceration	15	120 (85-140)	55	125 (90-185)
H pylori	30	125 (85-200)	40	135 (105-178)

TABLE 33
 Duodenal Blood Flow (Perfusion Units) of NSAID Patients
 Median (Interquartile Ranges) in the Presence of
 Smoking, Ulcers, or H pylori

	Present		Absent	
	Sample size	Value	Sample size	Value
Smoking	29	150* (100-175)	41	175 (135-250)
Duodenal Ulceration	12	123* (93-158)	68	160 (130-215)
H pylori	30	135* (100-175)	40	168 (140-245)

Significant drop compared with non-smokers or those without duodenal ulcers or H pylori:

* $p < 0.05$

10.5 DISCUSSION:

This study shows that, in patients treated with NSAID, duodenal blood flow is lowest in smokers and in those with duodenal ulcers or H pylori. Gastric blood flow does not seem to change significantly in the presence of any of these variables. Mucosal blood flow is greater in the duodenum than in the stomach, but no correlation could be found between mucosal blood flow and patients' age or type of arthritis. In the study group as a whole, the intake of NSAID is associated with lower mucosal blood flow in both the stomach and duodenum, compared with controls.

In addition to changes in cardiac output, there are several other factors that can influence the gastric mucosal microcirculation. Vasoconstriction has been observed to take place in the presence of vasopressin, catecholamines, angiotensin II, and after the activation of alpha-adrenergic receptors (213-216). Vasodilatation has been described with parasympathetic stimulation (215,217), gut peptides (gastrin, secretin, glucagon, etc) (209,213,217,218), and with most local metabolites such as histamine, serotonin, bradykinin, cyclic AMP, and prostaglandins (213,214,218,219). Gastric blood flow has also been found to change in response to drug intake: it was reduced by NSAID (14) and cimetidine (210) and increased (220) or unaffected (221) by misoprostol. Apart from NSAID, patients in this study were excluded if they took drugs such as cimetidine or misoprostol. It is worth noting, however, that the majority of the above studies (213-221) were carried out in laboratory animals, and when humans were included (210,211,220), duodenal blood flow and the effect of various demographic factors or H pylori were not investigated. Measurements made in the control group of this study confirm the general suppressive effects of NSAID on blood flow in

both the stomach and duodenum. The relatively small number studied does not allow for detailed analysis of the effect of H pylori or the demographic variables in the absence of NSAID, which lies beyond the scope of this study.

In this study, although NSAID patients older than 60 years tended to have lower mucosal blood flow than those younger than 40 years, this tendency was not statistically significant. This finding, alone, does not explain the higher incidence of NSAID related peptic ulcers or their complications previously reported in elderly people (95,145). Other factors should be considered, including the continuing increase in NSAID prescribing (95), the possible role of diet (222), and the effect of the cohort phenomenon (223).

Patients with NSAID-related gastric ulcers in this study, had similar gastric blood flow to those with normal gastric endoscopy. This is unlike the finding of a previous study (211) in which the presence of gastric ulceration was associated with lower gastric blood flow. The difference may have been related to the intake of NSAID: peptic ulcers in such cases can be related to multiple other mechanisms, including inhibition of both prostaglandin synthesis (7) and mucus secretion (17), and disruption of the gastric mucosal barrier (12). Alternatively, in the presence of low levels of gastric perfusion in NSAID patients, the method used in this study might not have been sensitive enough to detect small differences.

Smokers in the NSAID group had lower duodenal blood flow than non-smokers. This might explain, at least in part, the higher prevalence of ulcers in smokers, reported in previous studies

(42,75,224). Duodenal blood flow was also lower in the presence of *H pylori*, in this study. The reason for this is not clear, but it is interesting to note that recent in vitro studies carried out by the author (22,56) showed that *H pylori* culture filtrate intensified the inhibitory effects of indomethacin on two potential vasodilators: PGE₂ and cyclic AMP (213,214,219). However, multivariate analysis showed that the effects of smoking and *H pylori* infection could be accounted for by a reduction in blood flow only in those exposed to both of these risk factors, which might suggest a synergistic relationship between them.

The presence of duodenal ulceration in NSAID patients was associated with lower mucosal blood flow in the duodenum. This could be related to the high prevalence of smoking amongst duodenal ulcer patients. However, the independent association between duodenal ulceration and reduced duodenal blood flow should not be ignored as it approached statistical significance.

In the NSAID group as a whole, blood flow was lower in the gastric antrum than in the duodenum. The significance of such a finding is not clear but it could be relevant to understanding the tendency of NSAID related peptic damage to affect the antrum more frequently than the duodenum (5,42,Chapter 8) although it was not possible to demonstrate a correlation between gastric ulceration and gastric mucosal blood flow in this study.

In conclusion, duodenal blood flow in patients receiving NSAID appears to be lowest in smokers and in those with duodenal ulcers or

H pylori. The study could not demonstrate correlation with age, and gastric blood flow does not seem to be influenced by any of the above factors. These findings might be relevant to our understanding of NSAID related peptic damage. They also illustrate some differences between the gastric and the duodenal mucosa in their response to various risk factors in peptic ulcer disease.

SECTION VI
NSAID-RELATED ULCERS AND SECOND-LINE DRUGS

INTRODUCTION

The endoscopic study in Chapter 11 investigates the prevalence of peptic ulcers in patients taking NSAID only or with second-line drugs. Its finding of a lower prevalence of such ulcers in patients taking gold injections made it necessary to investigate the prevalence of both ulcers and H pylori in patients treated with gold or with another agent of similar anti-rheumatoid activity, such as sulphasalazine. This was justified on the basis of two main reasons: Firstly, the possible interaction between NSAID and H pylori (Chapter 2); and secondly, because of the historical evidence suggesting that gold compounds might have an antibacterial activity (Chapter 3).

CHAPTER 11

11 ENDOSCOPIC NON-STEROIDAL PEPTIC DAMAGE IN RHEUMATOID PATIENTS RECEIVING SECOND-LINE DRUGS

11.1 SUMMARY

Background: NSAID are commonly used together with second-line drugs for the control of rheumatoid arthritis. It is not clear whether an interaction exists between these agents with respect to peptic ulceration.

Aims: To assess NSAID-related gastric and duodenal abnormalities in patients treated with second-line drugs.

Methods: Using endoscopy, the prevalence of peptic ulceration was studied in 281 rheumatoid patients receiving NSAID alone, or in combination with second-line agents.

Results: Ulcers were found in 33 out of 96 patients who took NSAID only (33/96, 34%), compared with 8/33 (24%) on hydroxychloroquine, 10/30 (33%) on penicillamine, 13/46 (28%) on sulphasalazine, but only 11/76 (14%) on intramuscular gold plus NSAID ($X^2=7.95$, $0.001 < p < 0.01$, $p < 0.05$ using Bonferroni correction).

Conclusion: Second-line drugs do not seem to increase the prevalence of ulcers related to NSAID. On the contrary, fewer ulcers were found in patients receiving gold therapy, and this might have therapeutic implications.

11.2 INTRODUCTION

In rheumatoid arthritis, second-line drugs play an integral part in the treatment of this common and chronic form of arthritis. Recently, arguments have been put for the early, and therefore the more widespread use of these drugs, in an attempt to minimize the debilitating complications of rheumatoid disease (225,226). These benefits are often limited by the development of gastro-intestinal side effects, which are considered amongst the commonest causes of interruption or even the termination of second-line therapy (227,228). Previous studies have entirely relied on patients' symptoms in evaluating the gastro-intestinal side effects of second-line drugs (227,228). Since these agents are taken in combination with NSAID in the majority of cases, patients' symptoms might not reflect the true extent of peptic damage, which can be asymptomatic in many patients (81).

The aim of this study was to measure the prevalence of peptic injury in patients taking NSAID with or without second-line drugs, using endoscopy.

11.3 PATIENTS AND METHODS

Patients with adult onset rheumatoid arthritis were recruited provided they took NSAID for a minimum of one month and second-line therapy for at least 6 months, with the dose unchanged for 3 months or longer.

Second-line agents studied included intramuscular sodium aurothiomalate (gold), hydroxychloroquine, penicillamine and sulphasalazine. Patients with previous gastric surgery or those taking anti-ulcer therapy, steroids or cytotoxic agents were excluded.

Informed consent was obtained and endoscopy was performed after the administration of 5-15mg diazepam intravenously. The endoscopist was not aware of the details of patients' drugs.

Endoscopic findings were classified into normal, erosive changes, or ulcers. An ulcer was defined as a three dimensional punched out lesion of at least 5mm in diameter, and an erosion was a smaller and shallow bidimensional lesion.

Statistical analysis included Chi-square (X^2) test and Bonferroni correction for multiple comparisons.

11.4 RESULTS

Two hundred and eighty one patients were studied; they were re-grouped according to whether they took NSAID with or without second-line drugs.

The endoscopic findings are shown in Figure 15. The intake of gold plus NSAID appears to be associated with a significantly smaller number of ulcers than in any other group.

The patients' demographic details are shown in Table 34. Subjects were comparable in their ages except for those receiving

hydroxychloroquine: they tended to be younger than the rest. Female/male ratio was greater than 2:1 in all groups. However, there was a greater preponderance of females in patients taking hydroxychloroquine or penicillamine. The smoking and drinking habits were similar in the various groups.

The types of NSAID taken by various patients are shown in Table 35. Indomethacin, diclofenac, and naproxen were the most commonly used NSAID, being taken by approximately similar proportions of subjects in all groups. The dosage and frequency of NSAID intake were also similar amongst the various groups.

The presence or absence of indications for endoscopy are demonstrated in Table 36. The sulphasalazine group tended to have a greater proportion of cases with upper abdominal complaints. About a third of patients in other groups did not have anaemia or any complaints at all. These asymptomatic subjects still had a similar prevalence of peptic damage to that found in other subjects in their respective groups.

Patients with ulcers (75 in total) had a median age of 57 years and included 41 smokers. Ulcers were found in the gastric antrum in 49 cases, compared with 18 in the duodenum, 5 in the gastric body, and 3 in the oesophagus. A similar distribution of ulcers was found in various patients regardless of their therapeutic agents.

FIGURE 15

The endoscopic findings in rheumatoid patients receiving non-steroidal anti-inflammatory drugs with or without second-line drugs.

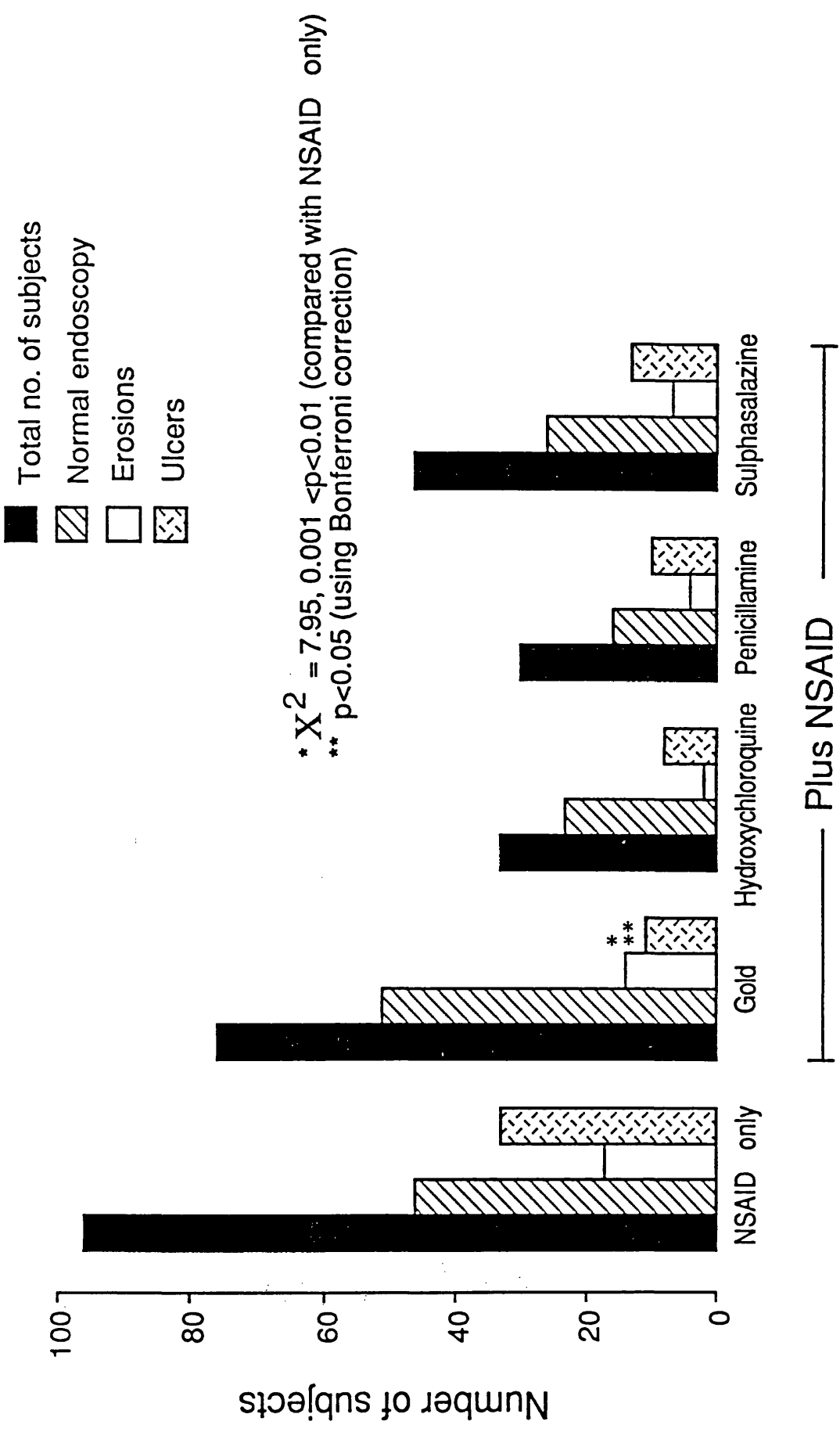


TABLE 34: Demographic Details of Patients in the Various Study Groups

	NSAID Only	Gold	Hydroxychloroquine	Penicillamine	Sulphasalazine
No of patients	96	76	33	30	46
Median Age (years)	59	57	46	57	57
Sex:					
Male	29	20	5	2	16
Female	67	56	28	28	30
Smokers	40	29	9	6	12
Alcohol drinkers	34	22	13	9	12

TABLE 35
Types of NSAID Used

Types of NSAID Used	NSAID Only (n=96)	Gold (n=76)	Hydroxychloroquine (n=33)	Penicillamine (n=30)	Sulphasalazine (n=46)
Indomethacin	20	24	5	6	11
Diclofenac	15	8	5	6	9
Naproxen	14	14	8	6	5
Ibuprofen	14	7	-	1	3
Flurbiprofen	9	5	5	6	5
Fenpufen	7	4	-	-	1
Ketoprofen	6	5	4	1	4
Piroxicam	6	3	2	2	4
Others	5	6	4	2	4

Table 36
The Presence or Absence of Indications for Endoscopy

Indication	Number of patients (%)			
	NSAID Only (n=96)	Gold (n=76)	Hydroxychloroquine (n=33)	Sulphasalazine (n=46)
Abdominal symptoms	37 (39%)	24 (32%)	9 (27%)	22 (48%)
Anaemia	30 (31%)	32 (42%)	9 (27%)	17 (37%)
Nil	29 (30%)	20 (26%)	15 (46%)	7 (15%)

11.5 DISCUSSION

This endoscopic study shows that the prevalence of peptic ulcers is lower in patients receiving gold plus NSAID, but unchanged in patients taking NSAID plus other second-line drugs in comparison with the intake of NSAID only.

There were unequal numbers of patient in the various groups. This is because patients were recruited at random, provided they satisfied the inclusion and exclusion criteria. In addition, patients receiving hydroxychloroquine tended to be younger than their counterparts in other groups. This reflects a management policy that involves giving hydroxychloroquine to young adults with rheumatoid arthritis, in preference to other second-line drugs (229,230). Despite being younger, they still had a similar prevalence of peptic ulcers to those taking NSAID only. This finding does not necessarily disagree with the epidemiological evidence of a high incidence of ulcer complications in the elderly (95,145) as the assessment of such complications lies beyond the scope of this study.

The nature of this study makes it difficult to assess patients' compliance with the intake of various drugs, especially if these drugs had to be taken for relatively long periods of time. This is further emphasized by the insistence that second-line drugs had to be administered for a minimum of six months before patients could be included in the study, as the disease modifying activity of these agents may take several months to manifest itself (225-233). It could be argued that patients receiving gold therapy developed fewer ulcers because their arthritis was better controlled and, as a

result took NSAID less frequently. This is unlikely to be the case, given the fact that several studies have demonstrated that the anti-rheumatoid efficacy of gold is comparable to that of sulphasalazine (227,228,232).

Although it is somewhat difficult to explain the lower prevalence of peptic ulcers in association with gold therapy, comparison has recently been made with bismuth, which is classified very close to gold in the periodic table of elements (101). The bactericidal activity of gold was 30 times stronger than that of bismuth in *H pylori* cultures (101). Colloidal bismuth subcitrate was also found to have a protective effect against aspirin-induced gastric microbleeding, and this protection occurred despite suppression of mucosal prostaglandin production by aspirin (33). Whether gold has a similar protective activity remains a matter of speculation. Although, the precise role of *H pylori* in mediating NSAID-induced damage is not fully clear, it is interesting to note that the combination of indomethacin and *H pylori* culture filtrate has recently been shown to reduce gastric mucosal viability and its prostaglandin E_2 concentration to a greater extent than indomethacin alone, in vitro (22) and that NSAID patients infected with *H pylori* have a higher prevalence of ulcers than patients without such infection (Chapters 2,7). The effect of gold on the stomach is probably different from that on the colonic mucosa where it may cause colitis (108).

Previous studies investigating the possible gastro-intestinal side-effects of second-line agents in the presence of NSAID have largely relied on patients' symptoms (227,233). This endoscopic

study suggests that the intake of hydroxychloroquine, penicillamine or sulphasalazine does not increase the prevalence of peptic ulcers in patients on NSAID. This is of particular relevance to patients receiving sulphasalazine: their tendency to develop abdominal complaints does not necessarily indicate a higher prevalence of peptic ulceration, and might be due to a central effect especially in slow acetylators (234).

In conclusion, second-line agents, other than gold, do not seem to alter the prevalence of NSAID-related peptic damage. The apparently beneficial effect of gold might have some therapeutic implications.

CHAPTER 12

12 NSAID-RELATED ULCERS AND H PYLORI IN RHEUMATOID ARTHRITIS PATIENTS RECEIVING GOLD OR SULPHASALAZINE

12.1 SUMMARY

Background: The conflicting reports on gold and H pylori could be related to the use of serological tests of unproven value in NSAID patients, and to the lack of the appropriate control groups. More importantly, the endoscopic consequences of the possible effect of gold on H pylori have not been investigated.

Aims: To assess the prevalence of H pylori and peptic ulcers in rheumatoid patients being treated with gold sodium thiomalate plus NSAID, sulphasalazine plus NSAID, or NSAID only.

Patients and Methods: Eighty-five patients receiving treatment for at least six months were endoscoped and H pylori studied in gastric antral biopsies by both culture and histology. Endoscopic abnormalities were classified into ulcers (measuring 5mm in diameter or more) and erosions (smaller lesions).

Results: H pylori (and ulcers) were found in 17 (12) of 31 patients on NSAID only and 21 (9) of 27 patients on sulphasalazine plus NSAID compared with 9 (3) of 27 patients receiving gold plus NSAID, $p < 0.05$, analysis of variance.

Conclusion: Patients treated with gold and NSAID had the lowest prevalence of detectable H pylori. This could explain the apparent reduction in the prevalence of peptic ulcers in this group and, if confirmed in larger randomised studies, might have therapeutic implications.

12.2 INTRODUCTION

Nearly a century after its description by Koch (106), the antimicrobial activity of gold salts is being reconsidered because of its potential use in the treatment of H pylori. Recent in vitro studies have suggested that bactericidal effect of gold sodium thiomalate was 30 times stronger than bismuth against H pylori cultures (101), and that such effect could be related to the inhibition of H pylori urease activity by gold (102). The author has found that rheumatoid arthritic patients treated with intramuscular gold were less likely to have NSAID-related ulcers compared with patients treated with other second-line drugs (82, Chapter 11). However, H pylori status was not known in Chapter 11, and endoscopy was not performed in recent serological studies, which showed conflicting results on the prevalence of H pylori in patients with gold salts (103,104).

The aim of this study was to assess the prevalence of H pylori and peptic ulcers in patients receiving gold plus NSAID compared with those treated with sulphasalazine plus NSAID, or NSAID only.

12.3 PATIENTS AND METHODS

Patients with adult rheumatoid arthritis, according to the criteria of the American Rheumatism Association (149), were recruited from the Rheumatology Out-Patient Clinic provided they had been taking NSAID only or with intramuscular gold or sulphasalazine for at least six months. Sulphasalazine was chosen as a control agent because its second-line activity has been recognised as being comparable to that of gold (228,232). Patients were excluded if they took ulcer healing drugs, antibiotics, steroids, cytotoxic drugs, or if they had a history of previous gastric surgery. Arthritic pain was assessed on a 4 point scale: 0(no pain), 1(mild), 2(moderate), and 3 (severe) joint pain.

Informed consent was obtained and endoscopy performed using midazolam 4-8mg intravenously for sedation. Endoscopic abnormalities were classified into ulcers (3 dimensional punched out lesions measuring at least 5mm in diameter) or erosions (smaller bidimensional lesions (82,Chapter 11). Gastric antral biopsies were taken to check for H pylori by both histology and culture, as previously described (61).

Statistical analyses involved the analysis of variance or the Kruskal-Wallis test, where appropriate. The endoscopist, histologist, and bacteriologist were not aware of the details of patients medications and the study was approved by the local Ethics Committee.

12.4 RESULTS

Ninety patients were studied, five of whom were excluded

because of uncertainty about recent antibiotic use. The remaining 85 patients entered the final analysis and included 31 on NSAID only, 27 on NSAID plus sulphasalazine, and 27 on NSAID plus intramuscular gold. The duration of second-line therapy was 2(1-2) years for sulphasalazine and 2 (1.5-6) years for gold, median (interquartile ranges).

The prevalence of detectable H pylori, ulcers, and erosions is demonstrated in Figure 16. Patients in the gold group had the lowest prevalence of both detectable H pylori and ulcers, but no difference could be found in the number of erosions amongst the three study groups. Ulcers diagnosed in this study were distributed as follows: 7 gastric (and 5 duodenal) in patients taking NSAID only, 4(5) in the sulphasalazine group, and only 3 gastric ulcers in patients receiving gold injections. The differences in the prevalence of duodenal ulcers were not statistically significant. All gastric ulcers affected the antrum, and were found to be benign by histology. Erosions were also more likely to affect the antrum than the duodenum, in a ratio of 2:1, in all groups.

The demographic characteristics of patients are shown in Table 37. The smoking and drinking habits were comparable amongst the groups, but patients treated with gold tended to be older.

The duration and activity of rheumatoid arthritis are described in Table 38. The median duration of arthritis was greater than seven years in all groups, with a tendency to being longer in

patients treated with gold. Also, despite minor differences in their values, the parameters of disease activity measured in this study suggest that patients stabilised on the various long-term drugs had a similar degree of disease control at the time of endoscopy.

The details of NSAID used are presented in Table 39. No significant differences could be found in the type, dosage, or duration of NSAID used by patients in the three study groups. It must be emphasised that the duration of NSAID therapy shown in Table 39 refers to agents currently being taken, and that patients have been taking other NSAID for the duration of their arthritis, presented in Table 38.

FIGURE 16

Prevalence of H pylori, ulcers, and erosions

*: $p < 0.05$ (analysis of variance), compared with patients taking NSAID only or with sulphasalazine

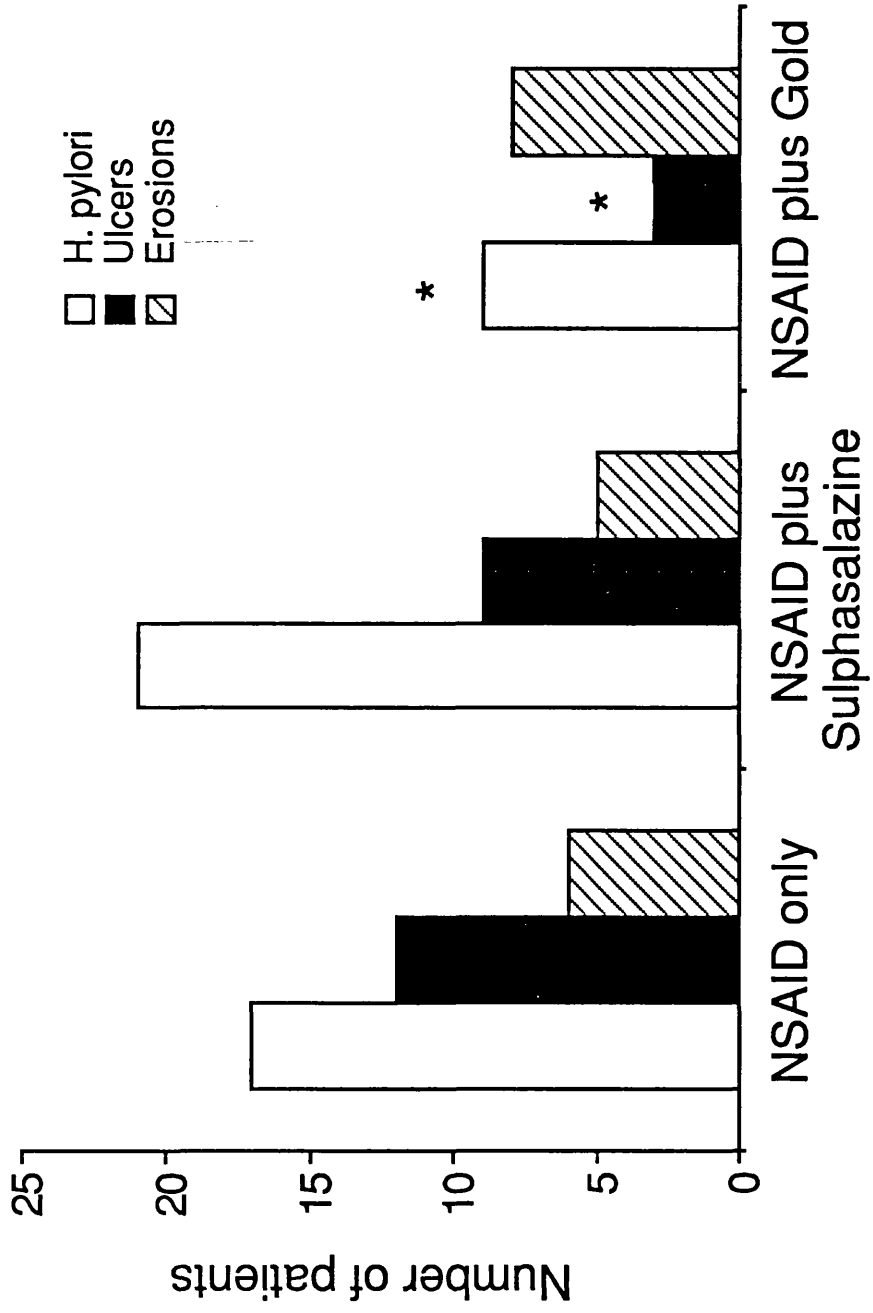


TABLE 37

Demographic Characteristics of the Study Groups

	NSAID only	NSAID plus Sulphasalazine	Gold
Number	31	27	27
Males	5	12	7
Females	26	15	20
Age (years) Median	55	51	60
(interquartile ranges)	(45-60)	(40-62)	(49-68)
Smokers	11	12	8
Drinkers	10	15	12
Patients with abdominal complaints	14	9	13

TABLE 38

Duration and Activity of Rheumatoid Arthritis

Median (interquartile ranges)

	NSAID only	NSAID plus Sulphasalazine	Gold
Duration (years)	7 (4-11)	7 (4-13)	11 (5-15)
Joint pain*	1 (2-3)	2 (1-2)	1 (1-2)
ESR (mm/hour)	30 (20-54)	27 (10-41)	23 (9-52)
CRP (mg/l)	16 (10-27)	21 (10-43)	17 (10-39)

* Grades of joint pain - 0: none, 1: mild, 2: moderate, and 3: severe

Table 39

Details of NSAID Used

Type:	NSAID plus		
	NSAID only	Sulphasalazine	Gold
Indomethacin	7	5	7
Naproxen	6	3	4
Ketoprofen	6	-	4
Diclofenac	2	7	1
Fenbufen	3	2	2
Piroxicam	3	1	1
Nabumetone	1	4	-
Ibuprofen	-	3	1
Flurbiprofen	-	2	3
Others	3	2	4
Dose*, +	2(2-3)	3(2-3)	2(2-3)
Duration (years) +	2(1-4)	3(2-5)	2(2-4)

* Dosage scale - 1: minimal, 2: submaximal, and 3: maximal

+ Median (interquartile ranges)

12.5 DISCUSSION

This study suggests that patients receiving long-term NSAID and gold injections have a low prevalence of detectable H pylori, which in turn might explain the apparent reduction of the prevalence of peptic ulcers in this group.

Patients treated with gold tended to be older than those treated with sulphasalazine or NSAID only; this could explain the relatively longer duration of arthritis in the gold group. Several studies have shown that elderly patients are at a greater risk of having H pylori infection (96,136), and developing NSAID related peptic ulcers or their complications (95,145). It is interesting, therefore, to find that patients receiving gold plus NSAID still had a lower prevalence of H pylori and ulcers despite their tendency to be older.

Despite having fewer ulcers, patients in the gold group had as many abdominal complaints as those in the other groups. This illustrates the poor correlation between symptoms and peptic ulcers in patients treated with NSAID (81,82, Chapter 4). Also, patients in the sulphasalazine group did not have more gastrointestinal symptoms, which could be due to the possibility that they have become adapted to sulphasalazine by the time their endoscopy was performed (234).

The low prevalence of detectable H pylori in chronic gold and NSAID users could be due to a direct bactericidal effect (101), inhibition of H pylori urease activity by gold (102) or both. Another mechanism which has to be considered is the possibility that long-term intake of gold injections might indirectly influence the

survival of *H pylori* by its immunomodulatory activity; immunocompetent patients are more likely to be infected by *H pylori*, and this chronic infection may be due to occupation of an immunoprivileged site, and associated with interference with the host's cellular immune response (109,110). However, it is difficult to understand why such interesting observations should not apply to patients treated with sulphasalazine, which is known to have second-line activity comparable to that of gold (228,232). Chronic suppression, rather than eradication of *H pylori* by gold could also explain the smaller number of cases of detectable organisms in biopsies taken from long term gold users. This implies that *H pylori* might recur or become more easily detectable upon withdrawal of gold, although such possibility has not been studied before. The presumed effect of intramuscular gold on *H pylori* in this study is thought to be due to the distribution of gold into the gastric mucosa, gastric secretions, or both. Gastric tissue or juice levels of gold are not known in this study, but it could be speculated that such levels might have been higher if oral gold preparations were used, which in turn might be more effective against *H pylori*. The use of the currently available oral gold compounds is, however, limited by their tendency to cause diarrhoea, and by the relatively lower anti-rheumatoid efficacy in comparison with intramuscular gold.

These results are in agreement with another serological study (104) which also found a low prevalence of *H pylori* in rheumatoid arthritic patients treated with gold compounds. However, conflicting findings were reported by another study (103), and this could be due to the methodology of detecting active *H pylori* infection. The value of serological tests (103,104) has not been established in

patients receiving NSAID; such patients might have low prevalence of *H pylori* (23,75,83,84,Chapter 7) and this in turn might lower the specificity of the serological tests (78, Chapters 4,5). It is believed that these possibilities have been covered by studying multiple biopsy specimens using both culture and histology in this study.

There was no significant difference in the prevalence of endoscopic abnormalities amongst the study groups when the size of such lesions was not taken into consideration. However, patients treated with gold had the smallest number of ulcers measuring at least 5mm in diameter. The significance of finding superficial erosions in chronic NSAID users is not certain: there is no evidence that they progress to larger lesions or to suggest that they may lead to serious complications such as perforation. It is speculated that the low prevalence of ulcers in the gold group might be related to the relative rarity of detectable *H pylori* in this group. This is supported by the recent findings which suggest that NSAID related mucosal damage and peptic ulcers are more likely to develop in the presence of *H pylori* (23,78, Chapters 2,7).

Another interesting finding in this study is the absence of duodenal ulcers in patients treated with gold. *H pylori* is typically associated with duodenal ulceration, and the low prevalence of these organisms in gold treated patients could explain, at least in part, the lack of duodenal ulcers in such patients. The differences in the numbers of duodenal ulcers amongst the study groups did not, however, reach statistical significance, and it is worth noting that the interaction between NSAID and *H pylori* is not the only mechanism by which NSAID cause mucosal damage (Chapters 1,2).

In conclusion, patients receiving long term NSAID plus gold have lower prevalence of detectable H pylori than those treated with NSAID only or NSAID plus sulphasalazine. This could explain the smaller number of peptic ulcers found in the gold group, and might have therapeutic implications, if confirmed by larger randomised studies.

SECTION VII
CONCLUSIONS, AND REFERENCES

CONCLUSIONS

This Thesis has addressed several aspects of the pathogenesis of NSAID-related peptic ulcer disease, including the interaction between NSAID and H pylori, the effect of NSAID on the efficacy of H pylori diagnostic tests, the histological features of NSAID-related mucosal damage, the influence of NSAID and H pylori on mucosal blood flow, and the interaction between NSAID and anti-rheumatoid second-line drugs. The main conclusions of these studies are as follows:

DIAGNOSTIC STUDIES

NSAID do not affect the sensitivity and specificity of the three biopsy-related tests used for the diagnosis of H pylori (culture, histology, and CLO-test).

Amongst the four serological tests studied, Bio-Rad GAP test IgG has the highest sensitivity but the lowest specificity, regardless of NSAID intake, compared with Bio-lab Malakit, Helico-G, and Pyloriset tests. The specificity of all the serological tests was also lower in patients taking NSAID.

The prevalence of NSAID-related peptic ulcers is higher in patients with debilitating arthritis, as assessed by the Health Assessment Questionnaire and endoscopy. The use of this questionnaire might, therefore, help in the process of selecting patients for prophylactic therapy against NSAID-induced peptic damage.

HISTOLOGICAL STUDIES

In the absence of gastro-duodenal surgery and heavy alcohol intake, chemical gastritis is characteristic of chronic NSAID intake.

Although H pylori is relatively less common in the presence of NSAID, chemical gastritis and H pylori-related gastritis are associated with a higher prevalence of NSAID-related ulcers than in patients without either of these histological abnormalities.

A strong association exists between duodenal ulcers and active chronic duodenitis, gastric metaplasia in the duodenum, and H pylori-positive gastritis, regardless of NSAID intake.

Active chronic duodenitis is relatively rare in chronic NSAID users, and this might explain why duodenal ulcers occur less frequently than gastric ulcers in patients treated with NSAID.

Duodenal ulcers are still as common in NSAID patients as in those not treated with these drugs, which in turn highlights the multifactorial nature of the pathogenesis of NSAID-related damage.

The prevalence of endoscopic oesophagitis is higher in NSAID patients with upper abdominal complaints than in their symptomatic counterparts not receiving NSAID.

Papillary elongation and basal cell hyperplasia, the major features of histological oesophagitis, are, however, less common in patients treated with NSAID. This suggests that histological oesophagitis might not be critical for the development of

NSAID-related oesophageal ulcers or erosions, which is comparable to other cases of pill-related oesophageal damage.

BLOOD FLOW STUDIES

Both gastric and duodenal blood flow values are lower in patients treated with NSAID than those not taking these drugs

Duodenal, but not gastric, mucosal blood flow is lowest in chronic NSAID users who smoke, and those with H pylori or duodenal ulceration.

Step-wise linear regression analysis suggests that the combination of smoking and H pylori has an independent effect in lowering duodenal mucosal blood flow in patient taking NSAID.

Duodenal mucosal bleed flow, in general, is higher than that of the gastric antrum in patients receiving NSAID. Not unlike the rarity of active chronic duodenitis, these findings might explain why duodenal ulcers are less common than gastric antral ulcers in NSAID patients, and emphasize the role of other mechanisms of NSAID-related damage.

NSAID AND SECOND-LINE DRUGS

NSAID-related ulcers occur less commonly in patients receiving gold injections than those treated with NSAID only, or with sulphasalazine, penicillamine, and hydroxychloroquine.

The prevalence of both H pylori and peptic ulcers is lower in patients treated with gold injections plus NSAID compared with those

treated with NSAID only or with sulphasalazine. These results might influence the choice of second-line drugs for the control of rheumatoid arthritis especially in patients with a history of peptic ulcer disease.

REFERENCES

- 1 Flower RJ
Drugs which inhibit prostaglandin biosynthesis.
Pharmacol. Rev. 1974;26:33-67
- 2 Shen TY
The chemical and pharmacologic characteristics of sulindac. In
Current Concepts on Anti-inflammatory drugs. CM Platz (ed).
New York, Biomedical Information Corporation Publications, 1980,
p1
- 3 Maddox IS
The role of copper in prostaglandin production.
Biochem. Biophys. Acta 1973;306:74-81
- 4 Dvornik D
Etodolac: effect on prostaglandin concentrations in gastric
mucosa of rat.
Life Sci. 1985;36 1157-62
- 5 Taha AS, McLaughlin S, Holland PJ, Kelly RW, Sturrock RD, Russell
RI
Effect on gastric and duodenal mucosal prostaglandins of repeated
intake of therapeutic doses of naproxen and etodolac in
rheumatoid arthritis.
Ann.Rheum. Dis 1990;49:354-8
- 6 Main IH, Whittle BJ
Investigation of the vasodilator and anti secretory role of
prostaglandin in the rat gastric mucosa by use of non-steroidal
anti inflammatory drugs.
Br J Pharmacol 1975;53:217-24

- 7 Whittle BJR, Higgs GA, Eakins KE, Moncada S, Vane JR
Selective inhibition of prostaglandin production in inflammatory exudates and gastric mucosa
Nature 1980;284:271-3
- 8 Redfern JS, Lee E, Feldman M
Effects of indomethacin on gastric mucosal prostaglandins in humans. Correlation with mucosal damage.
Gastroenterology 1987;92:969-77
- 9 Robert A, Nezamis JE, Lancaster C, Hanchar AJ
Cytoprotection by prostaglandins in rats: prevention of gastric necrosis by alcohol, HCl, NaOH, hypertronic NaCl, and thermal injury
Gastroenterology 1979;77:433-43
- 10 Baskin W, Ivey KJ, Krause WJ, Jeffrey GE, Gemmell RT
Aspirin induced ultrastructural changes in human gastric mucosa: correlation with potential differences
Ann Intern Med 1976; 85:299-303
- 11 Rainsford KD, Willis C
Relationship of gastric mucosal damage induced in pigs by anti inflammatory drugs to their effect on prostaglandin production.
Dig Dis Sci 1982; 27:624-35
- 12 Cooke AR
The role of the mucosal barrier in drug induced gastric ulceration and erosions.
Dig Dis Sci 1976;21:155-64
- 13 Feldman M, Colturic TJ
Effect of indomethacin on gastric acid and bicarbonate secretion in humans
Gastroenterology 1984;87:1339-43

- 14 Gerkens JF, Shand DG, Flexner C, Nies A, Oates J, Data J
Effect of indomethacin and aspirin on gastric blood flow and acid secretion
J Pharmacol Exp Ther 1977;203:646-52
- 15 Levine RA, Schwartzel EH
Effect of indomethacin on basal and histamine stimulated human gastric acid secretion
Gut 1984;25:718-22
- 16 Menguy R, Masters YF
Effect of aspirin on gastric mucus secretion
Surg Gynaecol Obstet 1965;120:92-8
- 17 Menguy R, Desbaillets L
Role of inhibition of gastric mucous secretion in the phenomenon of gastric mucosal injury by indomethacin
Am J Dig Dis 1967;12:862-6
- 18 Saroseik J, Mizuta K, Slomiany A, Slomiany B
Effect of acetylsalicylic acid on gastric mucin activity, permeability to hydrogen ion, and susceptibility to pepsin
Biochem. Pharmacol 1986;35:4291-5
- 19 Rees WDW, Gribbons LC, Turnberg LA
Effects of non-steroidal anti inflammatory drugs and prostaglandins on alkaline secretion by rabbit gastric fundus in vitro
Gut 1983;24:784-9
- 20 Kent TH, Cardelli RM, Stanler FW
Small intestinal ulcers and intestinal flora in rats given indomethacin
Am J Pathol 1969;54:237-49

- 21 Robert A, Asano T
Resistance of germ-free rats to indomethacin-induced intestinal lesions
Prostaglandins 1977;14:333-41
- 22 Taha AS, Kelly RW, Gemmell CG, Lee FD, Russell RI
The interaction between Helicobacter pylori culture filtrate and indomethacin: effects on the integrity of human gastric antral mucosa and its prostaglandin E₂ production in vitro
Aliment Pharmacol Therap 1990;4:265-73
- 23 Taha AS, Nakshabendi I, Lee FD, Sturrock RD, Russell RI
Chemical gastritis and Helicobacter pylori-related gastritis in patients receiving non-steroidal anti inflammatory drugs. Comparison and correlation with peptic ulceration
J Clin Pathol 1992;45:135-9
- 24 Robert A, Schultz AJ, Nezamis JE, Lancaster C
Gastric antisecretory and anti-ulcer properties of PGE₂, 15-methyl-PGE₂ and 16, 16-dimethyl PGE₂. Intravenous, oral and intrajejunal administration
Gastroneterology 1976;70:359-70
- 25 Chen MC, Amirian DA, Toomey M, Sanders MJ, Soll AH
Prostanoid inhibition of canine parietal cells: mediation by the inhibitory guanosine triphosphate - binding protein of adenylate cyclase
Gastroenterology 1988;94:1121-9
- 26 Levine RA, Jyotirmoy N, King RL
Non-salicylate non-steroidal anti inflammatory drugs augment prestimulated acid secretion in rabbit parietal cells. Investigations of the mechanisms of action.
Gastroenterology 1991;101:756-65
- 27 Tarnawski A, Hollander D, Gergely H, Stachura J
Comparison of antacid, sucralfate, cimetidine and ranitidine in protection of the gastric mucosa against ethanol injury
Am J Med 1985; 79 (suppl 2c):19-23

- 28 Hollander D, Tarnawski A, Cummings D, Krause WJ, Gergely H, Zipser RD
Cytoprotective action of antacids against alcohol-induced gastric mucosal injury. Morphologic, ultrastructural, and functional time sequence analysis.
Gastroenterology 1984;86:A1114
- 29 Tarnawski A, Hollander D, Cummings D, Krause WJ, Gergely H, Zipser RD
Are antacids acid neutralizers only? Histologic, ultrastructural and functional changes in normal gastric mucosa induced by antacids
Gastroenterology 1984;86:A1276
- 30 Peskar BM, Hollander A, Peskar BA
Effect of carbenoxolone on prostaglandin synthesis and degradation
J Pharm Pharmac 1976;28:146-8
- 31 Peskar BM, Weiler H
Carbenoxolone inhibits thromboxane B₂ synthesis by human gastric mucosa
Gut 1983;24:
- 32 Capurso L, Koch MM, Tonon GC, Maladrino S
Tripotassium dicitrate bismuthate (TDB): Cytoprotection in rats
Gastroenterology 1983;86:A1040
- 33 Konturck SJ, Kwiecien N, Obtulowicz W et al
Effects of colloidal bismuth subcitrate on aspirin-induced microbleeding, DNA loss, and prostaglandin formation in humans
Scand J Gastroenterol 1988;23:861-6
- 34 Hollander D, Tarnawski A, Gergely H, Zipser RD
Sucralfate protection of the gastric mucosa against ethanol-induced injury: a prostaglandin-mediated process?
Scand J Gastroenterol 1984 (suppl 101);19:97-102

- 35 Quadros E, Ramsamooj E, Wilson DE
Relationship between sucralfate, gastric cytoprotection and
prostaglandin and mucus synthesis and secretion
Gastroenterology 1985;88:A1548
- 36 Tarnawski A, Hollander D, Krause WJ, Zipser RD, Strachura J,
Gergely H
Does sucralfate affect the normal gastric mucosa? Histologic,
ultrastructural and functional assessment of the rat
Gastroenterology 1986;90:893-905
- 37 Roth SE, Bennet RE, Mitche CS, Hartman RJ
Cimetidine therapy in non-steroidal anti inflammatory drug
gastropathy. Double-blind long term evaluation
Arch Intern Med 1987;147:1798-1801
- 38 Kimmey MB, Silverstein FE, Saunders DR, Chapman RC
Reduction of endoscopically assessed acute aspirin induced
gastric mucosal injury with cimetidine
Dig Dis Sci 1987;32:851-6
- 39 Ehsanullah RSB, Page MC, Tildesley G, Wood JR
Prevention of gastro-duodenal damage induced by non-steroidal
anti inflammatory drugs: controlled trial of ranitidine
Br Med. J 1988;297:1017-21
- 40 Lancaster-Smith MJ, Jaderberg ME, Jackson DA
Ranitidine in the treatment of non-steroidal anti inflammatory
drug associated gastric and duodenal ulcer
Gut 1991;32:252-5
- 41 Graham DY, Agrawal NM, Roth SH
Prevention of NSAID-induced gastric ulcer with misoprostol:
multicentre, double-blind, placebo-controlled trial
Lancet 1988;2:1277-80

- 42 Farah D, Sturrock RD, Russell RI
Peptic ulceration in rheumatoid arthritis
Ann Rheum Dis 1988;47:478-80
- 43 Lanza FL, Aspinal RL, Swabb EA, Davies RE, Rack MF, Rubin A
Double-blind, placebo-controlled endoscopic comparison of the
mucosal protective effects of misoprostol versus cimetidine on
tolmetin-induced mucosal injury to the stomach and duodenum
Gastroenterology 1988;95:289-94
- 44 Graham DY, Stromatt SC, Jaszewski R, White RH, Triadafilopoulos G
Prevention of duodenal ulcer in arthritics who are chronic NSAID
users: a multi center trial of the role of misoprostol
Gastroenterology 1991;100:A75
- 45 Smith JTL, Pounder RE, Nwokolo CU, Lanzon-Miller S, Evans DG,
Graham DY, Evans (Jr) DJ
Inappropriate hypergastrinaemia in asymptomatic healthy subjects
infected with Helicobacter pylori
Gut 1990;31:522-25
- 46 Marshall BJ, Armstrong JA, McGeachie DB, Glancy RJ
Attempt to fulfil Koch's postulates for pyloric Campylobacter
Med J Aust 1985;142:436-7
- 47 Morris A, Nicholson G
Ingestion of Campylobacter pyloridis causes gastritis and raised
fasting gastric pH
Am J Gastroenterol 1987;82:192-9
- 48 Peterson W, Lee E, Skoghund M
The role of Campylobacter pyloridis in epidemic gastritis with
hypochlorhydria
Gastroenterology 1987;92:A1575

- 49 Graham DY, Smith JL, Alpert LC, Yoshimura HH
Epidemic achlorhydria is not viral but is caused by *Campylobacter pyloridis*
Gastroenterology 1987;92:A1412
- 50 Cave DR, Vargas M
Effect of *Campylobacter pylori* protein on acid secretion by parietal cells
Lancet 1989;ii:187-9
- 51 Chittajallu RS, Harwood J, Dorrian CA, McColl KEL
Is *Helicobacter pylori* related hypergastrinaemia due to the bacterium inhibiting parietal cell function?
Gut 1991;32:A1206
- 52 Graham DY, Lew GM, Lechago J
Helicobacter pylori related increase in plasma gastrin: What is the status of the antral G cells in man?
Gastroenterology 1991;100:A75
- 53 Chittajallu RS, Dorrian CA, Neithercut WD, Dahill S, McColl KEL
Is *Helicobacter pylori* associated hypergastrinaemia due to the bacterium's urease activity or the antral gastritis?
Gut 1991;32:1286-90
- 54 Wyatt JI, Rathbone BJ, Green DM, Primrose J
Raised fasting serum gastrin in chronic gastritis is independent of *Campylobacter pylori* status and duodenal ulceration
Gut 1989;30:A1483
- 55 Teichmann RK, Pratschke E, Grab J, Hammer C, Brendel W
Gastrin release by interleukin-2 and gamma-interferon in vitro
Can J Physiol Pharmacol 1986;64 (suppl):62

- 56 Taha AS, Fraser WD, Kelly RW, Gemmell CG, Lee RD, Russell RI
Inhibition of human gastric cyclic AMP production by *Helicobacter pylori* protein - possible involvement of mucosal prostaglandin E₂
Aliment Pharmacol Therap 1991;5:379-89
- 57 De Witte TJ, Geerdink PJ, Lamers CB, Boerbooms AM, van der Korst JK
Hypochlorhydria and hypergastrinaemia in rheumatoid arthritis
Ann Rheum Dis 1979;38:14-17
- 58 Sarosiek J, Slomiany A, Slomiany BL
Evidence for weakening of gastric mucus integrity by *Campylobacter pylori*
Scand J Gastroenterology 1988;23:585-90
- 59 Morley J, Bray MA, Jones RW, Nugteren DM, Van Drop DA
Prostaglandin and thromboxane production by human and guinea pig macrophages and leucocytes
Prostaglandins 1979;17:730-6
- 60 Hawkey CJ
Synthesis of prostaglandin E₂, thromboxane B₂ and prostaglandin catabolism in gastritis and gastric ulcer
Gut 1986;27:1484-92
- 61 Taha AS, Boothman P, Holland P, McKinlay A, Upadhyay R, Kelly RW, Lee F, Russell RI
Gastric mucosal prostaglandin synthesis in the presence of *Campylobacter pylori* in patients with gastric ulcers and non-ulcer dyspepsia
Am J Gastroenterol 1990;85:47-50
- 62 Cryer B, Faust T, Goldschmiedt M, Redfern JS, Lee E, Feldman M
Mucosal prostaglandins in peptic ulcer disease: correlation with demographic and histologic parameters
Gastroenterology 1991;100:A49

- 63 Sievert W, Lambert JR, Peacock T, Nicholson L
Gastroduodenal mucosal prostaglandin synthesis: is there an association with Helicobacter pylori and chronic active gastritis?
Gastroenterology 1991;100:A161
- 64 Goren A, Fotherby KJ, Shorthouse M, Wight DGD, Hunter JO
Campylobacter pylori and acid secretion
Lancet 1989;ii:212-3
- 65 Gana TJ, MacPherson BR, Koo J
Gastric mucosal blood flow in misoprostol pretreated aspirin-induced ulceration
Ann Surg 1988;207:327-34
- 66 Arakawa T, Nakamura H, Chono S, Satoh H, Fukuda T, Saeki Y, Kobayashi K
Absence of effect of 16, 16-dimethyl prostaglandin E₂ on reduction of gastric mucosal blood flow caused by indomethacin in rats
Dig Dis Sci 1989;34:1369-73
- 67 Taha AS, Angerson W, Nakshabendi I, Beekman H, Morran C, Sturrock RD, Russell RI
Gastric and duodenal mucosal blood flow in patients receiving non-steroidal anti-inflammatory drugs - influence of age, smoking, ulceration and Helicobacter pylori
Aliment Pharmacol Therap 1993; 7:41-45
- 68 Rosam AC, Wallace JL, Whittle BJR
Potent ulcerogenic actions of platelet-activating factor on the stomach
Nature 1986;319:54-6
- 69 Denizot Y, Sobhau J, Rambaud JC, Lewin M, Thomas Y, Benveniste J
PAF-acether synthesis by H pylori
Gut 1990;38:1242-6

- 70 Wallace JL, Keenan CM, Granger N
Gastric ulceration induced by non-steroidal anti inflammatory
drugs is a neutrophil-dependent process
Am J Physiol 1990;259:G462-7
- 71 Sobala GM, King RFG, Axon ATR, Dixon MF
Reflux gastritis in the intact stomach
J Clin Pathol 1990;43:303-6
- 72 Dixon MF, O'Connor HJ, Axon ATR, King RFJG, Johnston D
Reflux gastritis: distinct histopathological entity?
J Clin Pathol 1986;39:524-30
- 73 Laine L, Marin-Sorensen M, Weinstein WM
The histology of gastric erosions in patients taking
non-steroidal anti inflammatory drugs (NSAIDs): a prospective
study
Gastroenterology 1988;94:A247
- 74 Graham DY, Smith JL, Spjut HJ, Torres E
Gastric adaptation: studies in humans during continuous aspirin
administration
Gastroenterology 1988;95:327-33
- 75 Martin DF, Montgomery E, Dobek AS, Patrissi GA, Peura DA
Campylobacter pylori, NSAIDs, and smoking: risk factors for
peptic ulcer disease
Am J Gastroenterol 1989;84:1268-72
- 76 Graham DY, Lidsky MD, Cox AM, Evans DJ, Evans DG, Alpert L, Klein
PD, Sessoms SL, Michaletz PA, Saeed ZA
Long-term non-steroidal anti inflammatory drug use and
Helicobacter pylori infection
Gastroenterology 1991; 100:1653-7

- 77 Lanza FL, Evans DG, Graham DY
Effect of Helicobacter pylori infection on the severity of
gastroduodenal mucosa injury after the acute administration of
naproxen or aspirin to normal volunteers
Am J Gastroenterol 1991;86:735-7
- 78 Taha AS, Boothman P, Nakshabendi I, Reid J, Morran C, Gemmel CG,
Lee FD, Sturrock RD, Russell RI
Diagnostic tests for Helicobacter pylori - comparison and
influence of non-steroidal anti inflammatory drugs
J Clin Pathol 1992;45:709-12
- 79 Sobala GM, Crabtree JE, Pentith JA, Rathbone JA, Shallcross TM,
Wyatt JI, Dixon MF, Heatley RV, Axon ATR
Screening dyspepsia by serology to Helicobacter pylori
Lancet 1991;ii:94-6
- 80 Graham DY
Helicobacter pylori and the endoscopist: Whether to diagnose
Gastro-intest Endosc 1991;37:577-9
- 81 Skander MP, Ryan FP
Non-steroidal anti inflammatory drugs and pain-free peptic
ulceration in the elderly
Br Med J 1988;297:833-4
- 82 Taha AS, Capell HA, Sturrock RD, Russell RI
Non-steroidal peptic damage in rheumatoid patients receiving
second-line drugs
Am J Gastroenterol 1991;86:1588-91
- 83 Caselli M, Pazzi P, La Corte R, Aleotti A, Trevisani L,
Stabellini G
Campylobacter-like organisms, non-steroidal anti inflammatory
drugs and gastric lesions in patients with rheumatoid arthritis
Digestion 1989;44:101-4

- 84 Laine L, Marin-Sorensen M, Weinstein WM
Helicobacter pylori (HP) prevalence and mucosal injury in gastric ulcers (GUs): relationship to chronic non-steroidal anti-inflammatory drug (NSAID) ingestion
Gastroenterology 1991;100:A103
- 85 Marshall BJ, McGeachie DB, Rogers PA, Glancy RJ
Pyloric Campylobacter infection and gastroduodenal disease
Med J Aust 1985;142:439-44
- 86 von Wulffen H, Grote HJ, Gatermann S, Loning T, Berger B, Buhl C
Immunoblot analysis of immune response to Campylobacter pylori and its clinical associates
J Clin Pathol 1988;41:653-9
- 87 Newell DG
Identification of the outer membrane proteins of Campylobacter pyloridis and antigenic cross reactivity between C pyloridis and C jejuni
J Clin Microbiol 1987;133:163-70
- 88 Apel I, Jacobs E, Kist M, Bredt W
Antibody response of patients against a 120kDa surface protein of Campylobacter pylori
Zbl Bakt Hyg A 1988;268:271-6
- 89 Crabtree JE, Mahony MJ, Taylor JD, Heatley RV, Littlewood JM, Tompkins DS
Immune responses to Helicobacter pylori in children with recurrent abdominal pain
J Clin Pathol 1991;44:768-71
- 90 Crabtree JE, Taylor JD, Wyatt HI, Heatley RV, Shallcross TM, Tompkins DS, Rathbone BJ
Mucosal IgA recognition of Helicobacter pylori 120 kDa protein, peptic ulcerations and gastric pathology
Lancet 1991;ii:332-5

- 91 Figura N, Guglielmetti P, Rossolini A, Barber A, Cusi G, Musmanno Ra, Russi M, Quaranta S
Cytotoxin production by *Campylobacter pylori* strains isolated from patients with peptic ulcers and from patients with chronic gastritis only
J Clin Microbiol 1989;27:225-6
- 92 Konturek SJ, Bilski J, Kwiecien N et al
De-Nol stimulates gastric and duodenal alkaline secretion through prostaglandin-dependent mechanisms
Gut 1987;27:1557-63
- 93 Bjarnason I, Hayllar J, Smethurst, Price A, Gumpel MJ
Metronidazole reduces intestinal inflammation and blood loss in non-steroidal anti inflammatory drug induced enteropathy
Gut 1992;33:1204-8
- 94 Baum C, Kennedy DL, Forbes MB
Utilization of non-steroidal anti inflammatory drugs
Arthritis rheum 1985;28:686-92
- 95 Walt R, Katchinski B, Logan R, Ashley L, Langman MJS
Rising frequency of ulcer perforations in elderly people in the United Kingdom
Lancet 1986;i:489-92
- 96 Graham DY, Klein PD, Opekun AR, Boutton TW
Effect of age on the frequency of active *Campylobacter pylori* infection diagnosed by the [13C] urea breath test in normal subjects and patients with peptic ulcer disease
J Infect Dis 1988;157:777-80
- 97 Epstein WV
Parenteral gold therapy for rheumatoid arthritis: a treatment whose time has gone
J Rheumatol 1989;16:1291-94

- 98 Epstein WV, Henke CJ, Yelin EH, Katz PP
Effects of parenterally administered gold therapy on the course
of adult rheumatoid arthritis
Ann Intern Med 1991;114:437-44
- 99 Corkhill MM, Kirkham BW, Chicanza IC, Gibson T, Panayi GS
Intramuscular depot methyl prednisolone induction of
chrysotherapy in rheumatoid arthritis: a 24 week randomized
controlled trial
Br Rheumatol 1990;29:274-79
- 100 Lehtinen K, Isomaki H
Intramuscular gold therapy is associated with long survival in
patients with rheumatoid arthritis
J Rheumatol 1991;18:524-29
- 101 Barga LL, Sarosiek J, Marshall B, McCallum RW, Barret L, Guerrant
R
Gold - a new potential drug for the treatment of Helicobacter
pylori infection
Gastroenterology 1990;98:A25
- 102 Sarosiek J, Roche JK, McCallum RW, Marshall B
Urease - gold sodium thiomalate complex in inhibition of urease
activity
Gastroenterology 1990;98:A119
- 103 Gubbins GP, Schubert TT, Attansasio F, Lubetsky M, Perez-Perez
GI, Blaser MJ
Gold and seroprevalence of Helicobacter pylori in rheumatoid
patients
Gastroenterology 1991;100:A76
- 104 Yu ZJ, Sarosiek J, Feng T, Marshall BJ, Kaplan P, Davis JS,
McCallum RW
Colonisation of gastroduodenal mucosa with Helicobacter pylori in
patients with rheumatoid arthritis during treatment with gold
compounds
Gastroenterology 1991;100:A190

- 105 Taha AS, Sturrock RD, Russell RI
Helicobacter pylori and peptic ulcers in rheumatoid arthritis
patients receiving gold, sulphasalazine and non-steroidal anti
inflammatory drugs
Am J Gastroenterol 1992; 87:1732-5
- 106 Koch R
Weitere Mitteilungen über ein Heilmittel gegen Tuberculose
Dtsch Med Wschr 1890;16:1029-32
- 107 Forestier J
L'Aurotherapies dans les rheumations chroniques
Bulletins et Memoires de la Societe Medicale des Hopitaux de
Paris 1929;44:323
- 108 Wright A, Benfield GFA, Felix-Davis D
Ischaemic colitis and immune complexes during gold therapy for
rheumatoid arthritis
Ann Rheum Dis 1984;43:495-7
- 109 Rathbone BJ, Wyatt JI, Heatley V
Immunological aspects of Campylobacter pylori infection
Eur J Gastroenterol 1989;1:13-16
- 110 Logan RHP, Polson RJ, Rae G et al
Helicobacter pylori and HIV infection
Lancet 1990;i:1456
- 111 Warren J, Marsall B
Unidentified curved bacilli on gastric epithelium in active
chronic gastritis
Lancet 1983;i:1273-5

- 112 Price A, Levi J, Dolby J, Dunnscombe P, Smith A, Clark J, Stephenson M
Campylobacter pyloridis in peptic ulcer disease: microbiology, pathology and scanning microscopy
Gut 1985;26:1183-8
- 113 Cohan J, Humphries H, Dooley C, Keane C, Gilligan D, McKenna D, Sweeney E, O'Morain C
Campylobacter pylori and recurrence of duodenal ulcers - a 12 month follow up
Lancet 1987;ii:1109-11
- 114 Marshall B, Warren J, Blincow E, Philips M, Goodwin CS, Murray R, Blackbourn SJ, Waters TE, Sanderson CR
Prospective double-blind trial of duodenal ulcer relapse after eradication of Campylobacter pylori
Lancet 1988;ii:1437-42
- 115 Rauws EA, Tytgat GN
Cure of duodenal ulcer associated with eradication of Helicobacter pylori
Lancet 1990;i:1233-5
- 116 Jones D, Lessells A, Eldridge J
Campylobacter-like organisms on the gastric mucosa: culture, histological and serological studies
J Clin Pathol 1984;37:1002-6
- 117 Goodwin C, Blincow E, Warren J, Sanderson C, Easton L
Evaluation of cultural techniques for isolating Campylobacter pyloridis from endoscopic biopsies of gastric mucosa
J Clin Pathol 1985;38:1127-31
- 118 Hazell S, Borody T, Lee A
Campylobacter pyloridis gastritis: detection of urease as a marker of bacterial colonisation and gastritis
Am J Gastroenterol 1987;82:292-6

- 119 Evans DJ, Evans DG, Graham DY, Klein PD
A sensitive and specific serologic test for detection of
Campylobacter pylori infection
Gastroenterology 1989;96:1004-8
- 120 Ching CK
Screening for Helicobacter pylori in dyspepsia
Lancet 1991;ii:394
- 121 Westblom TU, Madan E, Czinn S, Gudipati S, Midkiff BR
Sensitivity and specificity of Pyloriset, a latex agglutination
test for detection of Helicobacter pylori infection
Gastroenterology 1991;100:A184
- 122 Whitehead R, Truelove SC, Bear MWL
The histological diagnosis of chronic gastritis in fiberoptic
gastroscope biopsy specimens
J Clin Pathol 1972;25:1-11
- 123 Dixon MF, Wyatt J, Burk DA, Rathbone BJ
Lymphocytic gastritis - relationship to Campylobacter pylori
infection
J Pathol 1988;154:125-32
- 124 McNemar Q
Note on the sampling error of the difference between correlated
proportions or percentages
Psychometrika 1947;12:152-7
- 125 Morris A, Ali MR, Brown P, Lane M, Patton K
Campylobacter pylori infection in biopsy specimens of gastric
antrum; laboratory diagnosis and estimation of sampling error
J Clin Pathol 1989;42:727-32

- 126 Meyer B, Werth B, Beglinger C, Dill S, Drewe J, Vischer WA, Eggers RH, Bauer FE, Stadler GA
Helicobacter pylori infection in healthy people: a dynamic process?
Gut 1991;32:347-50
- 127 Vaira D, Miglioli M, Holton J, Vergura M, Modugno V, Nanetti A, Barbara L
Prevalence of IgG, IgA, IgM to Helicobacter pylori in 1018 blood donors
Gut 1991;32:A564
- 128 Caruso I, Bianchi Porro GB
Gastroscopic evaluation of anti inflammatory agents
Br Med J 1980;280:75-8
- 129 Jones STM, Clague RB, Eldridge J, Jones DM
Serological evidence of infection with Helicobacter pylori may predict gastrointestinal intolerance to non-steroidal anti inflammatory drug (NSAID) treatment in rheumatoid arthritis.
Br J Rheumatol 1991;30:16-20
- 130 Veenedall RA, Pena AS, Meijer JL, Endtz H Ph, van der Est MMC, van Duijn W, Eulderink F, Kreuning J, Lamers CBHW
Longterm serological surveillance after treatment of Helicobacter pylori infection
Gut 1991;32:1292-4
- 131 Das SS, Karim QN, Easmon CSF
Opsonophagocytosis of Campylobacter pylori
J Med Microbiol 1988;27:125-30
- 132 Goodwin CS, Blincow E, Paterson G et al
Enzyme-linked immunosorbent assay for Campylobacter pyloridis: correlation with presence of C pyloridis in the gastric mucosa
J Infect Dis 1987;155:488-94

- 133 Steer HW, Hawtin PR, Newell DG
An ELISA technique for the serodiagnosis of *Campylobacter pyloridis* infection in patients with gastritis and benign duodenal ulceration
Serodiag Immunotherp 1987;1:253-9
- 134 Perez-Perez GI, Dworkin BM, Chodos JE, Blaser MJ
Campylobacter pylori antibodies in humans
Ann Int Med 1988;109:11-7
- 135 Crabtree JE, Shallcross TM, Heatley RV, Wyatt JI
Evaluation of a commercial ELISA for serodiagnosis of *Helicobacter pylori* infection
J Clin Pathol 1991;44:326-8
- 136 Jones DM, Eldridge JA, Fox AJ, Sethi P, Whorrell PH
Antibody to the gastric *Campylobacter*-like organism "*Campylobacter pylorides*" - clinical correlation and distribution in the normal population
J Med Microbiol 1986;22:57-62
- 137 Horan MA, Fox RA
Ageing and the immune response - a unifying hypothesis?
Mechanisms of Ageing and Development 1984;26:165-81
- 138 Fox JG, Correa P, Taylor NS, Zavala D, Fontham E, Janney F, Rodriguez E, Hunter F, Diavolitis S
Campylobacter pylori-associated gastritis and immune response in a population at risk of gastric carcinoma
Am J Gastroenterol 1989;84:775-81
- 139 Karnes WE, Samloff JM, Sisurala M, Kekki M, Sipponen P, Kim SWR, Walsh JH
Positive serum antibody and negative tissue staining for *Helicobacter pylori* in subjects with atrophic body gastritis
Gastroenterology 1991;101:167-74

- 140 Perez-Perez GI, Blaser MJ
Conservation and diversity of *Campylobacter pyloridis* major
antigens
Infect Immun 1987;55:1256-63
- 141 Rathbone BJ, Wyatt JI, Worsley BW, Shires SE, Trejdosiewicz LK,
Heatley RV, Losowsky MS
Systemic and local antibody responses to gastric *Campylobacter*
pyloridis in non-ulcer dyspepsia
Gut 1986;27:642-7
- 142 Sugiyama T, Imai K, Yoshida H, Takayama Y, Yabana T, Yokota K,
Oguma K, Yachi A
A novel enzyme immunoassay for serodiagnosis of *Helicobacter*
pylori infection
Gastroenterology 1991;101:77-83
- 143 Megraud F, Axelsson C, Bouchard S, Camon C
Comparison of kits to detect circulating *Helicobacter pylori* IgG
antibodies
Gastroenterology 1991;100:A123
- 144 Duggan JM, Dobson Annette J, Johnson H, Fahey P
Peptic ulcer and non-steroidal anti inflammatory agents
Gut 1986;27:929-33
- 145 Sommerville K, Faulkner G, Langman M
Non-steroidal anti inflammatory drugs and bleeding peptic ulcer
Lancet 1986;i:462-4
- 146 Fries JF, Spitz PW, Kraines RG, Holman HR
Measurement of patient outcome in arthritis
Arthritis Rheum 1980;23:137-45

- 147 Sherrer YS, Bloch DA, Mitchell DM, Young DY, Fries JF
The development of disability in rheumatoid arthritis
Arthritis Rheum 1986;29:494-500
- 148 Kirwan JR, Reedback JS
Stanford Health Assessment Questionnaire modified to assess
disability in British patients with rheumatoid arthritis
Br J Rheumatol 1986;25:206-9
- 149 Arnett FC, Edworthy SM, Bloch DA et al
The American Rheumatism Association 1987 revised criteria for the
classification of rheumatoid arthritis
Arthritis Rehum 1988;31:315-24
- 150 Shield MJ, Fenn GC, Kiff P, Stead H, Gies S
Do differential rates of NSAID-associated gastro-duodenal damage
exist? - the results of a large cohort endoscopy study carried
out in RA and OA patients
Br J Rheumatol 1990;29(suppl):5
- 151 Editorial
Misoprostol: ulcer prophylaxis at what cost?
Lancet 1988;ii:1293-4
- 152 Drapanas T, Bethea M
Reflux gastritis following gastric surgery
Ann Surg 1974;79:618-27
- 153 Laine L, Weinstein WM
Histology of alcoholic haemorrhagic "gastritis": a prospective
evaluation
Gastroenterology 1988;94:1254-62

- 154 Kauffman GL, Aures D, Grossman MI
Intravenous indomethacin and aspirin reduce basal gastric mucosal
blood flow in dogs
Am J Physiol 1980;238:G131-4
- 155 Gear MWL, Truelove SC, Whitehead R
Gastric ulcer and gastritis
Gut 1972;12:639-45
- 156 Greenlaw E, Sheahan DG, De Luca V, Millder D, Myerson D, Myerson
P
Gastroduodenitis - broader concept of peptic ulcer disease
Dig Dis Sci 1980;25:660-72
- 157 Sipponen P, Seppala K, Aarynen M, Helska T, Kettunen P
Chronic gastritis and gastro-duodenal ulcer: a case control study
on risk of coexisting duodenal or gastric ulcer in patients with
gastritis
Gut 1989;30:922-9
- 158 Dixon MF, Sobala GM
Gastritis and duodenitis: the histopathological spectrum
Eur J Gastroenterol & Hepatol 1992; 4 (suppl.2) :S17-S23
- 159 Sipponen P
Longterm consequences of gastroduodenal inflammation
Eur J Gastroenterol & Hepatol 1992; 4 (suppl.2) :S25-S29
- 160 Upadhyay R, Howatson A, McKinaly A, Danesh BJZ, Sturrock RD,
Russell RI
Campylobacter pylori associated gastritis in patients with
rheumatoid arthritis taking non-steroidal anti inflammatory drugs
Br J Rheumatol 1988;27:113-6

- 161 McNulty CAM, Watson CM
Spiral bacteria of the gastric antrum
Lancet 1984;i:1068-9
- 162 Joffe SN, Lee FD, Blumgart LH
Duodenitis
Clin Gastroenterol 1978;7:635-50
- 163 Hasan M, Sircus W, Ferguson A
Duodenal mucosal architecture in non-specific and ulcer
associated duodenitis
Gut 1981;22:637-41
- 164 Wyatt JI, Rathbone BJ, Dixon MF, Heatley RV
Campylobacter pyloridis and acid induced gastric metaplasia in
the pathogenesis of duodenitis
J Clin Pathol 1987;40:841-8
- 165 Carrick J, Lee A, Hazell S, Ralston M, Daskalopoulos G
Campylobacter pylori, duodenal ulcer, and gastric metaplasia:
possible role of functional heterotropic tissue in ulcerogenesis
Gut 1989;30:790-7
- 166 Wyatt JI, Rathbone BJ, Sobala GM, Shallcross T, Heatley RV, Axon
ATR, Dixon MF
Gastric epithelium in the duodenum: its association with
Helicobacter pylori and inflammation
J Clin Pathol 1990;43:981-6
- 167 James AH
Gastric epithelium in the duodenum
Gut 1964;5:285-94

- 168 Patrick WJA, Denham D, Forrest APM
Mucosa change in the human duodenum: a light and electron
microscopic study and correlation with disease and gastric acid
secretion
Gut 1974;15:767-76
- 169 Rhodes J
Experimental production of gastric epithelium in the duodenum
Gut 1964;5:454-8
- 170 Winklestein A
Inflammation of the lower third of the oesophagus
JAMA 1935;104:906-9
- 171 Richter JE, Castell DO
Gastro-oesophageal reflux: pathogeneis, diagnosis and therapy
Ann Intern Med 1982;97:93-103
- 172 Dodds WJ, Dent J, Hogan WJ, Helm JF, Hauser R, Patel GK, Egide MS
Mechanisms of gastro-oesophageal reflux in patients with reflux
oesophagitis
N Engl J Med 1982;307:1547-52
- 173 Johnson KE, Ask P, Boryd B, Fransson SG, Tibbling L
Oesophagitis, signs of reflux and gastric acid secretion in
patients with symptoms of gastro-oesophageal reflux disease
Scand J Gastroenterol 1986;21:837-47
- 174 Lillemore KF, Johnson LF, Harmon JW
Role of the components of the gastroduodenal contents in
experimental acid oesophagitis
Surgery 1982;92:276-84

- 175 Goldberg HI, Dodds WJ, Gee S, Montgomery C, Zboraske FF
Role of acid and pepsin in acute experimental oesophagitis
Gastroenterology 1969;56:223-30
- 176 Cross FS, Wangenstein OH
Role of bile and pancreatic juice in production of oesophageal
erosions and anaemia
Exp Biol Med 1957;77:862-6
- 177 Kivilaakso E, Fromm D, Silen W
Effect of bile salts and related compounds on isolated
oesophageal mucosa
Surgery 1980;87:180-5
- 178 Lillemore KD, Johnson LF, Harmon JW
Alkaline oesophagitis: a comparison of the ability of components
of gastroduodenal contents to injure the rabbit oesophagus
Gastroenterology 1983;85:621-8
- 179 Stoker DL, Williams JG
Alkaline reflux oesophagitis
Gut 1991;32:1090-2
- 180 Kickendall JW, Friedman AC, Oyewole MA, Fleischer D, Johnson LF
Pill-induced oesophageal injury. Case reports and review of the
medical literature
Dig Dis Sci 1983;28:174-82
- 181 Bott S, Prakash C, McCallum RW
Medication-induced oesophageal injury: Survey of the literature
Am J Gastroenterol 1987;82:758-63

- 182 Ovarthlarnporn B, Kulwichit W, Hiranniramol S
Medication-induced oesophageal injury: report of 17 cases with
endoscopic documentation
Am J Gastroenterol 1991;86:748-50
- 183 Eng J, Sabanathan S
Drug-induced oesophagitis
Am J Gastroenterol 1991;86:1127-33
- 184 Wilkins WE, Ridley MG, Pozniak AL
Benign stricture of the oesophagus: role of non-steroidal anti
inflammatory drugs
Gut 1984;25:478-50
- 185 Bonoviana L, DeMeester TR, McChesney L, Schwizer W, Albertucci M,
Bailey RT
Drug-induced oesophageal strictures
Ann Surg 1987;206:173-83
- 186 Semble EL, Wu WC, Castell DO
Non-steroidal anti inflammatory drugs and oesophageal injury
Semin Arthritis Rheum 1989;19:99-109
- 187 Santucci L, Patoia L, Fiorucci S, Farroni F, Favero D, Morelli A
Oesophageal lesions during treatment with piroxicam
Br Med J 1990;300:1018
- 188 Minocha A, Greenbaum DS
Pill-oesophagitis caused by non-steroidal anti inflammatory drugs
Am J Gastroenterol 1991;86:1086-9

- 189 Aabakken L
Non-steroidal anti inflammatory drugs - the extending scope of
gastro-intestinal side effects
Aliment Pharmacol Therap 1991;6:143-62
- 190 Eastwood GL, Beck BD, Castell DO, Brown FC, Fletcher JR
Beneficial effect of indomethacin on acid-induced oesophagitis in
cats
Dig Dis Sci 1981;26:601-8
- 191 Northway MG, Eastwood GL, Libshitz HI, Feldman MS, Mamel JJ,
Szwarc IA
Anti inflammatory agents protect opossum oesophagus during
radiotherapy
Dig Dis Sci 1982;27:923-8
- 192 Ambrus JL, Ambrus CM, Lillie DB, Johnson RJ, Gastpar H, Kishel S
Effect of sodium meclofenamate on radiation-induced oesophagitis
and cystitis
J Med 1984;15:81-92
- 193 Tochner Z, Barnes M, Mitchell JB, Orr K, Glatstein E, Russo A
Protection by indomethacin against acute radiation oesophagitis
Digestion 1990;47:81-7
- 194 Stein BE, Schwarzman ML, Carrol MA, Stahl RE, Rosenthal WS
Role of arachidonic acid metabolites in acid-pepsin injury to
rabbit oesophagus
Gastroenterology 1989;97:278-83
- 195 Orlando RC
Oesophageal epithelial defence against acid injury
J Clin Gastroenterol 1991;13:S1-5

- 196 Savary M, Miller G
L'oesophagel Manuel et atlas d'endoscopie
Soleure: Gassmann 1977
- 197 Collins BJ, Elliot H, Sloan JM, McFarland RJ, Love AHG
Oesophageal histology in reflux oesophagitis
J Clin Pathol 1985;38:1265-72
- 198 Gear MWL, Wilkinson GP
Open access upper alimentary endoscopy
Br J Hosp Med 1989;41:438-44
- 199 Heller SR, Fellows IW, Ogilvie AL et al
Non-steroidal anti inflammatory drugs and benign oesophageal
stricture
Br Med J 1982;285:167-8
- 200 Barrison IG, Trewby PN, Kane SP
Oesophageal ulceration due to emepronium bromide
Endoscopy 1980;12:197-9
- 201 Robert A
Cytoprotection by prostaglandins
Gastroenterology 1979;77:761-7
- 202 Mukhopadhyay A, Ratten S, Goyal R
Effect of prostaglandin E2 on oesophageal motility in man
J Apple Physiol 1975;39:479-81

- 203 Dennish GW, Castell DO
Inhibitory effect of smoking on the lower oesophageal sphincter
N Eng J med 1971;284:1136-7
- 204 Khan TA, Shragge BW, Crispin JS, Lind JF
Oesophageal motility in the elderly
Am J Dig Dis 1977;22:1049-54
- 205 Kamada T, Sato N, Kawano S, Fusamoto H, Abe H
Gastric mucosal haemodynamics following thermal or head injury.
A clinical application of reflectance spectrophotometry
Gastroenterology 1982;83:535-40
- 206 Svanes K, Ulven A
Gastric ulceration associated with experimental vascular
occlusion
Digestion 1977;15:517-25
- 207 Whittle B, Kauffman GL, Moncada S
Vasoconstriction with thromboxane A₂ induces ulceration of the
gastric mucosa
Nature 1981;292:472-4
- 208 Harjola PT, Sivula A
Gastric ulceration following experimentally induced hypoxia and
haemorrhagic shock: in vitro study of pathogenesis in rabbits
Ann Surg 1966;163:21-8
- 209 Gerkens JF, Flexner C, Oates JA, Shand DG
Prostaglandin and histamine involvement in the gastric
vasodilator action of pentagastrin
J Pharmacol Exp Ther 1977;201:421-6
- 210 Lunde OC, Kvernebo K, Larsen S
Effect of pentagastrin and cimetidine on gastric blood flow
measured by laser Doppler flowmetry
Scand J Gastroenterol 1988;23:151-7
- 211 Lunde OC, Kvernebo K

- Gastric blood flow in patients with gastric ulcer measured by endoscopic laser Doppler flowmetry
Scand J Gastroenterol 1988;23:546-50
- 212 Johansson K, Ahn K, Lindhagen J, Lundgren O
Tissue penetration and measuring depth of laser Doppler flowmetry in the gastrointestinal application
Scand J Gastroenterol 1987;22:1081-8
- 213 Jacobson E, Lanciault G
The gastrointestinal vasculature. In: Scientific basis of Gastroenterology, H L Duthie, Wormsley KG (eds)
Edinburgh, London and New York, Churchill Livingstone
1979, pp26-48
- 214 Guth PH
The gastric microcirculation and gastric mucosal blood flow under normal and pathological conditions. In: Progress in Gastroenterology, Vol 1, GBJ Glass (ed). New York and London, Grune and Statton 1968, pp323-347
- 215 Leonard AS, Long D, French LA, Peter ET, Wangersteen OH
Pendular pattern in gastric secretion and blood flow following hypothalamic stimulation - origin of stress ulcer?
Surgery 1964;56:109-20
- 216 Guth PH, Smith E
Escape from vasoconstriction in the gastric microcirculation
Am J Physiol 1975;228:1893-5
- 217 Martinson J
The effect of graded vagal stimulation on gastric motility, secretion and blood flow in the rat
Acta Physiol Scand 1965;65:300-309

- 218 Guth PH, Smith E
The effect of gastrointestinal hormones on the gastric
microcirculation
Gastroenterology 1976;71:435-8
- 219 Leung FW, Robert A, Guth PH
Gastric mucosal blood flow in rats after the administration of
16, 16-dimethyl prostaglandin E₂ at a cytoprotective dose
Gastroenterology 1985;88:1948-53
- 220 Sato N, Kawano S, Fukuda M, Tsuji S, Kamada T
Misoprostol-induced changes in gastric mucosal haemodynamics
Am J Med 1987;83:(1A)15-21
- 221 Gana TG, MacPherson BR, Koo J
Gastric mucosal blood flow in misoprostol pretreated
aspirin-induced ulceration
Ann Surg 1988;207:327-34
- 222 Sonnenberg A
Dietary salt and gastric ulcer
Gut 1986;27:1138-42
- 223 Sonnenberg A
Geographical and temporal variations in the occurrence of peptic
ulcer disease
Scand J Gastroenterol 1985;20[suppl.110]:11-24

- 224 Freidman BD, Seigelaub AB, Seltzer CC
Cigarettes, alcohol, coffee and peptic ulcer
N Eng J Med 1974;290:469-73
- 225 Wilske KR, Healey LA
Remodeling the pyramid - A concept whose time has come
J Rheumatol 1989;16:565-7
- 226 Hess EV, Luggen ME
Remodeling the pyramid - A concept whose time has not yet come
J Rheumatol 1989;16:1175-6
- 227 Grindulis KA, McConkey B
Outcome of attempts to treat rheumatoid arthritis with gold,
penicillamine, sulphasalazine or dapsone
Ann Rheum Dis 1984;43:398-401
- 228 Pullar T, Hunter JA, Capell HA
Sulphasalazine in rheumatoid arthritis: a double blind comparison
of sulphasalazine with placebo and sodium aurothiomalate
Br Med J 1983;287:1102-4
- 229 Scull E
Chloroquine and hydroxychloroquine therapy in rheumatoid
arthritis
Arthritis Rheum 1962;5:30-6
- 230 Bunch TW, O'Duffy D, Tompkins RB, O'Fallon WM
Controlled trial of hydroxychloroquine and D-penicillamine singly
and in combination in the treatment of rheumatoid arthritis
Arthritis Rheum 1984;27:267-76

- 231 Research Sub-committee of the Empire Rheumatism Council
Gold therapy in rheumatoid arthritis: final report of multicentre
controlled trial
Ann Rheum Dis 1961;20:315-34
- 232 Bax ED, Amos RS
Sulphasalazine: a safe, effective agent for prolonged control of
rheumatoid arthritis. A comparison with sodium aurothiomalate
Ann Rheum Dis 1985;44:194-8
- 233 Kean WF, Dwosh IL, Anastassiades TP, Ford PM, Kelly HG
The toxicity pattern of D-penicillamine therapy. A guide to its
use in rheumatoid arthritis
Arthritis Rheum 1980;23:158-64
- 234 Pullar T, Hunter JA, Capell HA
Sulphasalazine in the treatment of rheumatoid arthritis:
Relationship of dose and serum levels to efficacy
Br J Rheumatol 1985;24:269-76

SECTION VIII

PUBLICATIONS RELATED TO THIS THESIS

EDITORIALS AND REVIEWS

- Taha AS, Russell RI
Mucosal damage by non-steroidal anti inflammatory drugs:
re-assessment of the mechanisms and implications for management
Gastroenterology today 1991;1:92-6
- Taha AS, Russell RI
Helicobacter pylori and non-steroidal anti inflammatory drugs -
uncomfortable partners in peptic ulcer disease
Gut 1993; 34: 580-83
- Taha AS, Sturrock RD
The gold-plated stomach?
Ann Rheum Dis 1993; 52: 89-90

ORIGINAL ARTICLES

- Taha AS, Boothman P, Nakshabendi I, Reid J, Morran C,
Gemmell CG, Lee FD, Sturrock RD, Russell RI
Diagnostic tests for Helicobacter pylori - comparison and
influence of non-steroidal anti inflammatory drugs
J Clin Pathol 1992;45:709-12
- Taha AS, Reid J, Boothman P, Gemmell CG, Lee FD, Sturrock
RD, Russell RI
Serological diagnosis of Helicobacter pylori - evaluation of four
tests in the presence or absence of non-steroidal anti
inflammatory drugs
Gut 1993;34: 461-65
- Taha AS, Morran C, Sturrock RD, Russell RI
The Health Assessment Questionnaire as a predictor of
non-steroidal peptic ulceration
Br J Rheumatol 1993;32: 135-38

- Taha AS, Nakshabendi I, Lee FD, Sturrock RD, Russell RI
Chemical gastritis and Helicobacter pylori-related gastritis in patients receiving non-steroidal anti inflammatory drugs - a comparison and correlation with peptic ulceration.
J Clin Pathol 1992;45:135-9
- Taha AS, Dahill S, Nakshabendi I, Lee FD, Sturrock RD, Russell RI
Duodenal histology, ulceration, and Helicobacter pylori in the presence or absence of non-steroidal anti inflammatory drugs
Gut 1993;34:1162-6
- Taha AS, Dahill S, Nakshabendi I, Lee FD, Sturrock RD, Russell RI
Non-steroidal anti-inflammatory drugs and the oesophagus - Endoscopic and histological studies
(under review)
- Taha AS, Angerson W, Nakshabendi I, Beekman H, Morran C, Sturrock RD, Russell RI
Gastric and duodenal mucosal blood flow in patients receiving non-steroidal anti inflammatory drugs - influence of age, smoking, ulceration and Helicobacter pylori
Aliment Pharmacol Therap 1993; 7:41-45
- Taha AS, Capell HA, Sturrock RD, Russell RI
Non-steroidal peptic damage in rheumatoid patients receiving second-line drugs
Am J Gastroenterol 1991;86:1588-91
- Taha AS, Sturrock RD, Russell RI
Helicobacter pylori and peptic ulcers in rheumatoid patients receiving gold, sulphasalazine, and non-steroidal anti inflammatory drugs
Am J Gastroenterol 1992; 87:1732-5

ABSTRACTS

Taha AS, Dahill S, Nakshabendi I et al

Lack of papillary elongation and basal cell hyperplasia of the oesophageal mucosa in patients receiving non-steroidal anti inflammatory drugs

Gut 1992;33 [suppl 2] T176:S44

Taha AS, Sturrock RD, Russell RI

Helicobacter pylori and peptic ulcers in rheumatoid arthritis patients receiving non-steroidal anti inflammatory drugs and gold or sulphasalazine

Gastroenterology 1992;102:A172

Taha AS, Sturrock RD, Russell RD

Diagnosing Helicobacter pylori in patients receiving non-steroidal anti inflammatory drugs - comparison of culture, histology, Helico G nd Pyloriset tests

Gastroenterology 1992;102:A172

Taha AS, Sturrock RD, Russell RI

Helicobacter pylori and peptic ulcers in rheumatoid arthritis patients receiving non-steroidal anti inflammatory drugs and gold or sulphasalazine

Gut 1992:33 [suppl 1] W33:S9

Taha AS, Sturrock RD, Russell RI

Gastric and duodenal mucosal blood flow in patients receiving non-steroidal anti inflammatory drugs

Br J Rheumatol 1992:31 [suppl 1] A54:27

Taha AS, Sturrock RD, Russell RI

The Health Assessment Questionnaire as a predictor of non-steroidal peptic ulceration

Br J Rheumatol 1992:31 [suppl 1] A53:27

- Hudson N, Taha AS, Sturrock RD, Russell RI, Hawkey CJ
The influence of *Helicobacter pylori* colonisation on
gastro-duodenal ulceration in patients on non-steroidal anti
inflammatory drugs
Gut 1992;33 [suppl 1] T165:S42
- Taha AS, Nakshabendi I, Boothman P, Russell RI
Chemical gastritis and *Helicobacter pylori* in patients receiving
non-steroidal anti inflammatory drugs
Gut 1992;32:A563-4
- Taha AS, Capell H, Sturrock RD, Russell RI
Does gold therapy reduce non-steroidal peptic damage in
rheumatoid arthritis?
Br J Rheumatol 1991;30 [suppl 2] A277:139
- Taha AS, Angerson W, Beekman H, Morran C, Hudson N, Hawkey
CJ, Sturrock RD, Russell RI
Arthritic joint dysfunction as a predictor of non-steroidal anti
inflammatory (NSAID) peptic ulceration
Gut 1991;32:A1221
- Taha AS, Morran C, Hudson N, Hawkey CJ, Sturrock RD, Russell
RI
Arthritic joint dysfunction as a predictor of non-steroidal anti
inflammatory (NSAID) peptic ulceration
Gut 1992;32:A1221
- Taha AS, Capell HA, Sturrock RD, Russell RI
Does gold therapy reduce non-steroidal peptic damage in
rheumatoid arthritis?
Gut 1991;32:A571
- Taha AS, Lee FD, Sturrock RD, Russell RI
Chemical gastritis and *Helicobacter pylori* in rheumatoid arthritis
patients receiving non-steroidal anti-inflammatory drugs -
correlation with peptic ulceration
Gastroenterology 1991;100:A171

PRESENTATION TO LEARNED SOCIETIES

Taha AS et al

Lack of papillary elongation and basal cell hyperplasia of the oesophageal mucosa in patients receiving non-steroidal anti inflammatory drugs

The British Society of Gastroenterology, Warwick 1992

Taha AS et al

Helicobacter pylori and peptic ulcers in rheumatoid arthritis patients receiving non-steroidal anti inflammatory drugs and gold or sulphasalazine

The American Gastroenterological Association, San Francisco 1992

Taha AS et al

Gastric and duodenal mucosal blood flow in patients receiving non-steroidal anti inflammatory drugs

The British Society for Rheumatology, Southampton 1992

Taha AS et al

The Health Assessment Questionnaire as a predictor of non-steroidal peptic ulceration

The British Society for Rheumatology, Southampton 1992

Taha AS et al

Serological diagnosis of Helicobacter pylori

V Workshop, European Group on Gastrointestinal Pathology and Helicobacter pylori. Dublin, Ireland 1992

Taha AS et al

Helicobacter pylori and peptic ulcers in rheumatoid arthritis patients receiving non-steroidal anti inflammatory drugs and gold or sulphasalazine

The British Society of Gastroenterology, Sheffield 1992

Taha AS et al

Does gold therapy reduce non-steroidal peptic damage in rheumatoid arthritis?

The British Society for Rheumatology, London 1991

Taha AS et al

Interference with duodenal mucosal blood flow by *Helicobacter pylori* in patients receiving non-steroidal anti inflammatory drugs
IV Workshop, European Group on Gastroduodenal Pathology and
Helicobacter pylori, Bologna, Italy 1991

Taha AS et al

Gastric and duodenal mucosal blood flow in patients receiving
non-steroidal anti inflammatory drugs (NSAID) - Influence of
age, smoking, ulceration and *Helicobacter* (H) *pylori*
British Society of Gastroenterology, London 1991

Taha AS et al

Arthritic joint dysfunction as a predictor of non-steroidal
(NSAID) peptic ulceration
British Society of Gastroenterology, London 1991

Taha AS et al

Chemical gastritis and *Helicobacter pylori* in patients receiving
non-steroidal anti inflammatory drugs - correlation with peptic
ulceration
The American Gastroenterological Association, New Orleans 1991

Taha AS et al

Does gold therapy reduce non-steroidal peptic damage in
rheumatoid arthritis?
The British Society of Gastroenterology, Manchester 1991

Taha AS et al

Chemical gastritis and *Helicobacter pylori* in patients receiving
non-steroidal anti inflammatory drugs - correlation with peptic
ulceration.
The British Society of Gastroenterology, Manchester 1991

APPENDIX

Definitions used in this thesis:

Smokers:

Subjects currently taking more than one cigarette, cigar, or a roll of tobacco per day.

Alcohol drinkers:

Subjects consuming more than one unit of alcohol per week.

Abdominal complaints:

Nausea, vomiting, heartburn, flatulence, indigestion, dyspepsia, and/or abdominal pain.

Sensitivity:

The frequency of a positive test in all subjects with H pylori infection. It is calculated by dividing the number of true positives by the sum of true positives and false negatives.

Specificity:

The frequency of a negative test in those without the infection. It is calculated by dividing the number of true negatives by the sum of true negatives and false positives.

The diagnosis of the vast majority of chronic NSAID users is rheumatoid arthritis, unless indicated otherwise in this thesis.

All studies included in this thesis are covered by ethical approval granted by the Ethics Committee of Glasgow Royal Infirmary, as part of an ongoing assessment of the natural history and treatment of NSAID related peptic ulcer disease.

