Sympathetic activation: a mechanism for morphine induced pain and rises in liver enzymes after cholecystectomy?

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Abstract

In patients with biliary type pain after cholecystectomy, morphine often precipitates pain and may induce rises in plasma concentrations of liver enzymes because of exaggerated or prolonged rises in intrabiliary pressure. In this study, changes in plasma concentrations of catecholamines and histamine were determined after the administration of morphine in patients with and without a two-fold or greater rise in the plasma concentration of aspartate aminotransferase at four hours. Those showing rises in aminotransferase had higher concentrations of noradrenaline at 40 and 60 minutes after morphine and higher concentrations of dopamine at 40 minutes after morphine. The two groups had similar concentrations of adrenaline and histamine. Attempts to inhibit rises in aminotransferase after morphine by pretreatment with histamine, serotonin and α -receptor blockers were largely unsuccessful, although inhibition was observed with phenoxybenzamine in two of five patients. Higher plasma concentrations of noradrenaline and dopamine before and soon after induction of pain in patients showing rises in aminotransferase are consistent with sympathetic activation but heterogeneity appears to exist in the response to α -receptor blockade.

An understanding of the events which follow the administration of morphine may provide clues to the pathogenesis of biliary type pain after cholecystectomy.¹⁻⁴ Instead of the expected analgesia, most patients report induction of pain at 45 to 60 minutes after intramuscular morphine which persists for one or two hours before settling spontaneously. At three hours after morphine, one-third of patients show rises in plasma concentrations of liver enzymes, particularly aminotransferase, and further rises are observed at four hours.² Rises in liver enzymes seem likely to reflect an exaggerated or prolonged rise in intrabiliary pressure as changes in enzymes are abolished by endoscopic sphincterotomy.^{2,3}

The effect of morphine on sphincter of Oddi motility involves two changes which may impede bile flow, an increase in the frequency of phasic contractions and an increase in the basal pressure.⁵ The former is antagonised by naloxone but the latter may be the result of a separate mechanism which is unrelated to mu receptors. Although morphine can have direct effects on smooth muscle,⁶ results from a variety of animal experiments raise the possibility that a major component of the action of morphine

involves indirect effects which could be mediated centrally by effects on autonomic activity^{7,8} or peripherally by release of serotonin⁹ or histamine.¹⁰ One of these mechanisms could be relevant to our recent observation of a higher frequency of abnormalities of sphincter motility in the subgroup of patients showing rises in aminotransferase after morphine.⁴

The purpose of this study was to evaluate metabolic responses to relatively low doses of morphine in patients with and without morphine induced rises in aminotransferase and to attempt to inhibit aminotransferase rises after morphine using drugs which antagonise the effects of histamine, serotonin, and noradrenaline.

Methods

PATIENTS

Thirty six adults with pain after cholecystectomy were included in the study. There were nine men and 27 women with a mean age of 48 years (range 19-67). All patients described episodes of pain in the epigastrium or right upper quadrant which radiated into the back and were often accompanied by nausea and vomiting.^{2,11} The mean interval since cholecystectomy was nine years and the mean duration of symptoms was five years. All patients had undergone extensive investigations including endoscopic retrograde cholangiopancreatography to exclude disorders such as bile duct stones, chronic pancreatitis, and peptic ulceration. The protocol was approved by the Ethics Committee at The Queen Elizabeth Hospital in 1985.

The morphine test^{2,4} was modified to permit blood collection for plasma concentrations of noradrenaline, adrenaline, dopamine, and histamine. Briefly, an intravenous needle was inserted between 08 00 h and 09 00 h and kept open with heparinised saline. Blood was taken after 10 and 20 minutes for basal concentrations of catecholamines and histamine. Thereafter, morphine was given intramuscularly at a dose of 0.12 mg/kg and further blood samples were taken after 20, 40, 60, 120, and 240 minutes. Additional blood samples were taken at 0, 180, and 240 minutes for plasma concentrations of liver enzymes and glucose. All tests were performed after an overnight fast and patients remained in bed and fasting for the duration of the test. The presence or absence of pain was recorded before each blood sample. At the end of the study, the overall pain experience was described and scaled as nil (0), discomfort only (one), mild pain (2), moderate pain (3), and severe pain (4). Patients were divided into two

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groups; those with rises in aminotransferase $>2\times N$ after four hours and those in whom concentrations of aminotransferase remained within the reference range.

Blood samples for catecholamines and histamine were collected in chilled heparinised tubes, placed in ice and centrifuged at 4°C. The plasma was stored at -20°C before assay in batches. Tubes for catecholamines contained glutathione (5 mmol/l) to prevent oxidation of adrenaline and noradrenaline. Plasma catecholamine concentrations were determined by a modification of the radioenzymatic technique described by Da Prada and Zurcher¹² while histamine concentrations were determined by a radioenzymatic method as described previously.¹³ Plasma concentrations of aminotransferase and glucose were determined by standard automated methods; the upper limit of the reference range for aminotransferase was 45 U/l. Rises in plasma aminotransferase to two-fold or more above the upper limit of the reference range were defined as abnormal and were expressed as $>2\times N^2$.

PRETREATMENT WITH DRUGS

Five patients with aminotransferase rises after morphine agreed to further studies after pretreatment with terfenadine, cyproheptadine, and phenoxybenzamine. These patients were included in the above protocol and had previously undergone at least one other morphine study which induced similar degrees of pain and resulted in similar rises in aminotransferase. In most instances, tests were performed at intervals of one week but the range was from three to 14 days. Terfenadine (120 mg) and phenoxybenzamine (20 mg) were given orally at two hours, and cyproheptadine (12 mg) at one hour before challenge with morphine. Blood was taken at 0, 180, and 240 minutes for plasma concentrations of liver enzymes. Degrees of pain were recorded throughout the study and, at the end of the study, patients were asked to compare the pain experience to that in previous studies.

STATISTICAL ANALYSIS

Mean age, mean interval since cholecystectomy, mean doses of morphine, mean rises in plasma glucose and mean pain scores in patients with and without rises in aminotransferase were compared by the Mann-Whitney U test. Plasma concentrations of noradrenaline, adrenaline, dopamine, and histamine were analysed using two-way analysis of variance (patients, times) for each amine and for each group separately. These values were used to calculate the least significant differences between any two means for significance levels of 5% based on a two-tailed *t* test. A

preliminary analysis of results after studies in 22 patients showed that those with rises in aminotransferase had significant rises in plasma concentrations of noradrenaline and dopamine but no change in the plasma concentration of histamine. In light of these results, additional patients were recruited for confirmation of changes in catecholamines but further histamine concentrations were not measured. After pretreatment with drugs, responses were considered abnormal if rises in the plasma concentration of aminotransferase were $<50\%$ or $>100\%$ of that in the control study. These arbitrary limits were based on our previous experience with 18 subjects having two or more morphine tests; in only one instance did the aminotransferase level during the second or subsequent test fall outside these limits.

Results

Results from three of 36 patients were excluded from statistical analysis. Two showed an intermediate rise in aminotransferase to levels above the reference range but $<2\times N$ while another showed a substantial rise in aminotransferase to 177 U/l but had plasma concentrations of noradrenaline at 0, 40, and 60 minutes after morphine of 6.28, 16.85, and 15.95 pmol/ml; concentrations which were substantially higher than all other patients. Of the remaining 33 patients, 16 showed rises in aminotransferase ($>2\times N$) while 17 had aminotransferase concentrations after four hours within the reference range. As shown in Table I, the two groups were well matched for age, interval since cholecystectomy, and dose of morphine. There were, however, more men showing rises in aminotransferase and this group had higher mean rises in plasma glucose ($p=0.0001$) and higher mean pain scores ($p=0.004$). Three patients were free of pain throughout the test, one with and two without rises in aminotransferase. Serial observations showed that the morphine injection did not result in a fall in blood pressure.

Plasma concentrations of noradrenaline, adrenaline, dopamine, and histamine in patients with and without rises in aminotransferase are shown in Table II. Those with rises in aminotransferase had significantly higher plasma concentrations of noradrenaline at 40 and 60 minutes after morphine and significantly higher concentrations of dopamine at 40 minutes after morphine. There were no significant differences in plasma concentrations of adrenaline or histamine.

Particular attention was directed to the relationship between rises in plasma concentrations of noradrenaline and the onset of pain in patients showing rises in aminotransferase. In

TABLE I Comparison of groups showing sex ratios, mean age, mean interval since cholecystectomy, mean dose of morphine, mean changes in plasma aminotransferase (AST), mean rise in plasma glucose over four hours and mean pain scores

	<i>n</i>	<i>F:M</i>	Mean age (yr)	Mean interval since surgery (yr)	Mean dose of morphine (mg)	Mean AST at 0 h (U/l)	Mean AST at 4 h (U/l)	Mean rise in pl glucose (mmol/l)	Mean pain score
AST $>2\times N$	16	8:8	51	8.8	8.1	23	179	1.9*	2.8†
AST $<2\times N$	17	17:0	44	9.0	8.3	21	25	0.6	1.7

* $p=0.0001$; † $p=0.004$.

TABLE II Mean plasma concentrations of noradrenaline, adrenaline, dopamine, and histamine before and after intramuscular morphine in patients with and without rises in aminotransferase (AST)

	'-10	'0	'20	'40	'60	'120	'240	LSD (5%)	RMS	df
Noradrenaline* (AST >2×N)	2.14	1.91	1.92	2.32‡	2.42‡	1.96	1.88	0.34	0.229	90
Noradrenaline* (AST <2×N)	1.59	1.57	1.56	1.48	1.54	1.59	1.54	0.22	0.108	96
Adrenaline* (AST >2×N)	0.27	0.18	0.15	0.27	0.28	0.30	0.24	0.14	0.037	90
Adrenaline* (AST <2×N)	0.25	0.20	0.17	0.20	0.19	0.12	0.19	0.10	0.018	96
Dopamine* (AST >2×N)	0.46	0.43	0.43	0.50‡	0.45	0.42	0.45	0.06	0.008	90
Dopamine* (AST <2×N)	0.40	0.41	0.42	0.41	0.40	0.40	0.41	0.06	0.008	96
Histamine† (AST >2×N)	0.45	0.32	0.36	0.33	0.34	0.28	0.24	0.16	0.034	60
Histamine† (AST <2×N)	0.32	0.29	0.34	0.40	0.38	0.37	0.27	0.17	0.037	60

LSD: least significant differences. RMS: residual mean square. df: degrees of freedom. *pmol/ml; †ng/ml; ‡p<0.05.

this group, eight were free of pain at 40 minutes while eight reported discomfort or mild pain. The presence of minor symptoms did not appear to influence noradrenaline concentrations as the mean rise in plasma noradrenaline between 20 and 40 minutes after morphine in patients without pain (0.45 pmol/ml) was similar to those with minor symptoms (0.32 pmol/ml). In all patients, the most severe symptoms were reported at either 60 or 90 minutes after morphine.

The effects of pretreatment with drugs on changes in aminotransferase after morphine are shown in the Figure. In general, basal concentrations of aminotransferase were reproducible although patient 4 had a raised concentration of aminotransferase on one occasion and also showed spontaneous changes in liver enzymes at other times. After pretreatment with terfenadine, higher concentrations of aminotransferase were noted in two patients. After cyproheptadine, concentrations of aminotransferase were higher in patient 2 and lower in patient 4. After phenoxybenzamine, concentrations of aminotransferase were higher in patient 2 and lower in patients 3 and 4. In all patients treated with phenoxybenzamine, the systolic blood pressure in the supine position fell by 10–20 mmHg. In individual patients, there was a close association between the degree of pain experienced with each test and the degree of rise of aminotransferase. After pretreatment with phenoxybenzamine, however, all patients experienced marked nausea, often with vomiting, between four hours and six hours after morphine.

Discussion

Although the relevance of responses to morphine has been debated,^{14 15} it was found that induction of pain and rises in plasma concentrations of aminotransferase are reproducible phenomena which are rarely observed in subjects who are asymptomatic after cholecystectomy.² Rises in intrabiliary pressure, presumably followed by distension of the biliary system, seem likely to be relevant to pain induced by morphine as the current study showed higher pain scores in patients with rises in aminotransferase and previous studies have shown that morphine induced pain was abolished or markedly attenuated by endoscopic sphincterotomy.^{2 3} The mechanism for pain in patients without rises in aminotransferase is unclear but may include lesser degrees of biliary distension, low thresholds for pain induced by changes in gastrointestinal motility and somatic manifestations of a variety of psychiatric disorders. Rises in aminotransferase are more prominent than rises in

other liver enzymes and appear to be the earliest marker of biliary obstruction.² Rises in plasma amylase are rare after morphine alone and appear to be less useful than aminotransferase as a marker of altered responsiveness to the combination of morphine and neostigmine.^{2 4}

Results from the present study showed that morphine induced significant rises in plasma concentrations of noradrenaline and dopamine in patients showing rises in plasma concentrations of aminotransferase. For noradrenaline, significant rises were noted at 40 and 60 minutes after morphine, times which often preceded and closely followed the onset of pain. For dopamine, differences between groups were statistically significant only at 40 minutes. Rises in plasma concentrations of noradrenaline and dopamine seem likely to reflect activation of the sympathetic nervous system and/or the adrenal medulla.¹⁶ The former may be more important as an examination of individual cases clearly showed rises in plasma noradrenaline before the onset of pain. In contrast, peak concentrations of adrenaline at 60 and 120 minutes coincided with maximal pain and may represent a secondary response rather than a significant event in the induction of symptoms. Although changes in plasma adrenaline did not reach statistical significance, rises in mean concentrations probably made a substantial contribution to rises in plasma glucose, perhaps in concert with other factors such as stimulation of glucagon secretion and inhibition of glucose clearance.⁸ Morphine usually decreases plasma concentrations of cortisol¹⁷ except in patients who experience severe pain where cortisol concentrations may rise. In the latter setting, there may be a more pronounced rise in plasma glucose (unpublished observations).

Although plasma concentrations of noradrenaline increased by only 27%, the degree of sympathetic activation seems likely to be much greater as only a fraction (perhaps 20%) of the noradrenaline released by sympathetic activity enters plasma.¹⁸ The majority is locally inactivated, largely by reuptake into sympathetic nerve endings. The mechanisms of sympathetic activation have not been addressed in this study but could include a low threshold for activation by morphine and other stimuli or a specific effect of morphine (and perhaps other opioids) on autonomic centres in the central nervous system.

Attempts to inhibit the effect of morphine by pretreatment with terfenadine, cyproheptadine and phenoxybenzamine were largely unsuccessful. After terfenadine, higher concentrations of aminotransferase (and more severe pain) were noted in two patients and raised the possibility

Plasma concentrations of aminotransferase at the time of administration of morphine and after four hours in five patients who had a control test and further studies after pre-treatment with terfenadine, cyproheptadine, and phenoxybenzamine. */**Studies in which rises in aminotransferase were >100% and <50% of values in the control study.

that the effect of morphine might be enhanced by histamine receptor blockade. If this observation can be confirmed by studies in additional patients, the possibility exists that histamine interacts with opioid or other receptors to modify the effects of morphine. After phenoxybenzamine, lower concentrations of aminotransferase were observed in two patients but the effect of morphine was abolished only in patient 4. Reasons for this heterogeneous response to phenoxybenzamine remain unclear but might include the inadequate blockade of α -receptors exposed to neuronal and/or circulating noradrenaline and the coexistence of opioid-mediated mechanisms which enhance sphincter activity but do not involve noradrenergic nerves.^{19,20}

The innervation of the sphincter of Oddi may provide an explanation for the characteristic biliary type pain induced by morphine. The region of the sphincter is richly supplied by noradrenergic nerve endings in various animals including primates,²¹⁻²³ particularly the circular muscle of the distal bile duct.²⁴ In experiments *in vivo*, sympathomimetic amines induced contraction and relaxation of sphincter tissue, apparently through effects on α - and β -receptors, respectively^{22,25} while in unpublished studies *in vitro*, we noted that smooth muscle from bovine sphincter of Oddi was more sensitive to noradrenaline than muscle from the cystic duct. If these observations apply in man, sympathetic activation may have a greater effect on sphincteric muscle than on intestinal muscle and may induce a greater rise in pressure in the bile duct after cholecystectomy than in other areas of the gastrointestinal tract.

The role of sympathetic activation by

morphine and other stimuli appears to warrant further investigation in patients with biliary type pain after cholecystectomy and perhaps in other functional bowel syndromes. Such patients could have high basal levels of sympathetic activity, low thresholds for sympathetic activation and/or enhanced reactivity of sphincter muscle to stimulation by catecholamines.²⁶ Sympathetic activation might also provide an explanation for the greater frequency of disordered sphincter motility in patients showing rises in aminotransferase,⁴ particularly if future studies show that α -receptor agonists are capable of increasing the sphincter basal pressure in man.

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