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Tryptophan-Morphine Interactions and Postoperative Pain

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FRANKLIN, K. B. J., F. V. ABBOTT, M. J. M. ENGLISH, M. E. JEANS, R. A. R. TASKER AND S. N. YOUNG. *Tryptophan-morphine interactions and postoperative pain.* PHARMACOL BIOCHEM BEHAV **35**(1) 157–163, 1990.—Patients undergoing abdominal surgery were infused with saline or the 5-hydroxytryptamine (5-HT) precursor tryptophan starting in the operating room and continuing for three hours in the recovery room. There was a nonsignificant trend for patients who received tryptophan to have higher pain scores. In the saline-treated patients, plasma tryptophan was below the range for normal healthy subjects, and there was a strong positive relationship between plasma tryptophan and morphine requirements. These data, taken together with animal data obtained using the formalin pain test, suggest that a 5-HT system in the brain can antagonize the dissociative state produced by morphine, which helps patients to tolerate pain. When plasma tryptophan falls below normal levels, brain 5-HT falls and morphine requirements are reduced. While tryptophan may potentiate spinal 5-HT function to decrease nociceptive afference in some circumstances, there may be clinical conditions in which the use of tryptophan is contraindicated.

Analgesia Morphine Tryptophan 5-HT Postoperative pain Humans

IN animal experiments morphine-induced antinociception is enhanced by increased serotonergic activity and reduced by decreased serotonergic activity (33, 34, 36). This, and anatomical evidence (6,7), indicate that a major component of morphine antinociception is mediated by the action of morphine on endogenous opioid receptors which increase 5-HT release in the spinal cord. Morphine is thought to stimulate opioid receptors located in the periaqueductal grey (39). These receptors modulate the activity of neurons which project to serotonin-containing neurons in the nucleus raphe magnus (28,39). Raphe magnus cells in turn innervate the spinal cord dorsal horn where release of 5-HT inhibits nociceptive afferent transmission (7). This is not the only mechanism of morphine antinociception (39), but the experimental evidence shows the raphe-spinal system to be a powerful inhibitor of nociceptive responses. However, while this theory of morphine antinociception is widely known and discussed in relation to clinical analgesia (21), there have been no systematic and experimentally controlled tests of its applicability to morphine analgesia in humans.

The synthesis of 5-HT is controlled in part by the availability of its precursor tryptophan in the brain (38). Under normal circumstances, tryptophan hydroxylase, the rate-limiting enzyme of the pathway to 5-HT, is only half saturated with tryptophan. This means that tryptophan loading can increase 5-HT synthesis up to, but no more than, two-fold (13,40). Thus, precursor loading is a convenient and ethically acceptable way of manipulating 5-HT synthesis and function in humans.

Tryptophan has been tested by itself as an analgesic in experiments on humans but the results have been inconsistent. Seltzer *et al.* (30) gave tryptophan or placebo to 30 normal subjects and looked at response to electrical stimulation of dental pulp. The threshold for perception of pain was not altered by tryptophan, but pain tolerance was significantly increased. Using a signal detection method, Lieberman (17) found that tryptophan reduced the discriminability of thermal pain stimuli. On the other hand, 14 days of tryptophan treatment (2 g per day) failed to alter radiant heat pain thresholds in groups of 20 female student volunteers (22).

Several studies have investigated tryptophan in relation to clinical pain with positive findings. King (16) gave tryptophan to five rhizotomy and cordotomy patients in whom pain had recurred and sensory deficits had diminished. With tryptophan their sensory deficits for both touch and pinprick reexpanded to the maximum extent initially recorded after surgery. In a double-blind study, Seltzer *et al.* (29) gave tryptophan or placebo to 30 patients with chronic maxillofacial pain. After four weeks there was a greater reduction in reported clinical pain and a greater increase in pain tolerance in the tryptophan group than the placebo group. Tryptophan (0.5 g every 6 hr) has also been reported to reduce pain 24 hr after endodontic surgery (31). Two studies reported negative results. One found tryptophan given at bedtime ineffective in patients with fibrositis syndrome (23), and the other found tryptophan or 5-hydroxytryptophan did not alter the pain of disc disease (24). The fact that tryptophan appears to reduce pain

sensitivity or increase pain tolerance in some clinical and experimental situations, and the neuropharmacological evidence that 5-HT is involved in the analgesic effect of opiates, suggest that tryptophan loading might reduce the amount of morphine required to control pain produced by surgery or trauma. To date there has been only one trial of tryptophan together with morphine which reported that tryptophan reinstated analgesia in 5 patients tolerant to morphine (14).

To examine the acute interaction of tryptophan and morphine in a controlled clinical situation, we have carried out a double-blind placebo-controlled study on the effect of tryptophan loading on the requirement for morphine in the recovery room during the first 4 hours following abdominal surgery.

METHOD

Subjects

Twenty-eight volunteers were selected from patients admitted for elective abdominal surgery (hysterectomy or cholecystectomy) at the Montreal General Hospital who met the criteria for American Surgical Association Grade I or II anesthesia risk. In addition, patients had to be able to speak and understand English and to have at least an 8th grade education. Potential subjects were excluded if they had standard biochemical tests outside the normal range, significant cardiovascular, respiratory or neurological disease, or if the patient was taking psychotropic medication. On the evening before surgery, the nurse research assistant, or one of the investigators, explained the study to the patients and those who agreed to participate were asked to sign a consent form approved by the Clinical Trials Committee of the hospital. Consent was confirmed the following morning before the patient entered the operating room.

Drugs and Randomization

Sterile solutions of normal saline or isotonic 1% L-tryptophan were prepared and packaged into 500 ml clear glass bottles and tested for purity, concentration, sterility and pyrogenicity by the Armand Frapier Institute of Montreal. The hospital pharmacist randomly assigned patients to the experimental or control group, and arranged for the appropriate bottles with coded labels to be sent to the operating room. Intravenous morphine was used for analgesia during the operation and in the recovery room.

Operating Room Procedure

For all patients, the anesthetist was one of the investigators (M.J.M.E.). No premedication was given. Anesthesia was induced with sodium thiopental 4 mg/kg IV and maintained with isoflurane and 100% O_2. Neuromuscular block was produced by pancuronium bromide (0.06 mg/kg) and reversed by pyridostigmine (0.14 mg/kg) and glycopyrrolate (0.006 mg/kg). For intraoperative analgesia, morphine was given in a loading dose of 8 mg followed by infusion at 2 mg/hr. The patient was fitted with a 24-ga arterial line in the nondominant hand and an intravenous line in the other hand. Blood was drawn from the arterial line for monitoring blood gasses and the concentrations of tryptophan and morphine. Tryptophan or placebo and morphine were infused through the IV line.

At the start of the operation the patient received a loading dose of tryptophan (10 mg/kg over 15 min) or an equivalent volume of saline. The infusion continued at a rate of 10 mg/kg per hour until the end of the study.

Recovery Room Procedure

In the recovery room the patient was attended continuously by the nurse research assistant who monitored vital signs, administered morphine, took blood samples and carried out the experimental pain assessment. When the patient was admitted to the recovery room and had regained consciousness, a blood sample (5 ml) was taken and the patient's pain was assessed with the pain rating scales. Thereafter, blood samples were taken every 15 min and pain was assessed every 30 min. Every five min the nurse checked whether the patient had pain, and if the patient required analgesic medication. If the patient requested medication for pain, an IV injection of morphine was given. The first 2 injections were of 4 mg and subsequent injections were 2 mg. Pain control was assumed to be achieved the first time the patient declined further medication. Subsequently, morphine (2 mg) was given after the 30-min pain assessments, if necessary, or if the patient spontaneously requested it. Vital signs were taken every 15 min and blood gases were measured 60 and 90 min after admission to the RR. When pain control was achieved and requests for morphine had been stable for 1 hour the study was terminated, the infusions were discontinued and, after 1 hr further observation, the patient was discharged to a ward. The amount of analgesic medication administered on the ward during the 3 days after the operation was recorded from the patient's chart. During this time, tablets of codeine/acetaminophen, 60/650 mg (Empracet™), or meperidine (Demerol™) were given. The total daily analgesic dose was calculated as equivalents of mg of meperidine by the formula: (mg of meperidine) + (number of Empracet tablets × 30).

Pain Assessment

Two measures of pain level were obtained. To assess the global pain level the patient was asked "Overall, how would you rate your pain right now on a scale where "0" is no pain and "5" is unbearable pain?"

Because pain quality may vary on several dimensions (20), patients were asked to describe their pain using 16 descriptors each associated with a specific quality of pain. These descriptors were derived from a multidimensional scaling analysis of the McGill Pain Questionnaire [Abbott unpublished; (35)]. The patient was asked to rate the pain descriptors on a scale where 0 represented "not at all" and 5 represented "very much." The descriptors were: sharp or stabbing, tearing, stinging or smarting, burning, cold or numb, throbbing, tight or squeezing, pressing, aching, cramping, spreading, penetrating, tender or sore, flickering, suffocating, sickening.

Biochemical Analyses

Total and free (nonalbumin bound) plasma tryptophan were measured by the fluorometric method of Denckla and Dewey (9). The free plasma tryptophan concentration was taken as the concentration in an ultrafiltrate of plasma prepared at 25° in an Amicon MPS-1 centrifugal ultrafilter using YMT membranes. Other large neutral amino acids in plasma were measured, in samples taken on entry to the recovery room, using an LKB Alpha Plus amino acid analyser.

To extract morphine from plasma, 0.5 ml plasma was added to 0.5 ml NH_4OH/HCl buffer (titrated to pH 9.0 with 2 N HCl) and 40 μl of 50 μg/ml naloxone as an internal standard. The mixture was placed on a 3 ml ChemElut 1001 column, washed on with 0.5 ml of the NH_4OH/HCl buffer and let stand for 5 min. Morphine was eluted with 7.5 ml of $CHCl_3$/n-butanol (90/10, v/v) and dried under nitrogen at 50°C. Morphine and naloxone were quantitated by HPLC using a Supelco C18 column and amperometric detection. The mobile phase was 75% 0.1 M KH_2PO_4, 15% methanol and 10% CH_3CN (v/v/v) and the oxidation potential was 690 mV

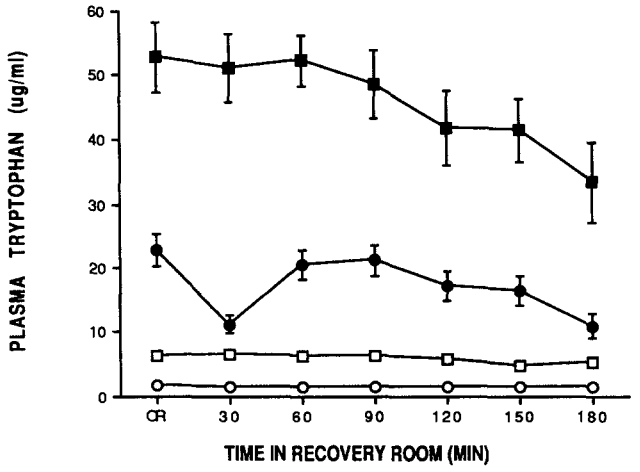

FIG. 1. Levels of plasma total tryptophan (square symbols) and plasma free tryptophan (round symbols) during 180 min in the recovery room for patients infused with tryptophan (filled symbols) or saline (open symbols). Vertical bars are standard errors.

relative to an Ag/AgCl electrode. Morphine was detected at 3.4 min and naloxone at 6.2 min. Morphine concentrations were read from a standard curve on which morphine concentration (10–200 ng/ml) was plotted against the morphine/naloxone ratios recorded from standards that had been passed through the extraction procedure.

RESULTS

The data from two patients were dropped from the analysis. In one patient pain was not controlled after 32 mg of IV morphine and she was discharged to the ward. The other patient was dropped from the study when it was discovered that her wound had been infiltrated with the local anesthetic bupivacaine. Complete data were obtained on 11 saline-infused patients and 15 tryptophan-infused patients. No patients received blood transfusion.

For both tryptophan-infused and control subjects, total and free plasma tryptophan were maximal in the operating room and fell steadily during the hours in the recovery room, $F(5,115) = 11.69$, $p<0.001$; $F(5,120) = 7.32$, $p<0.0001$ (Fig. 1).

Thirty min after the start of infusion the plasma total tryptophan level in tryptophan-infused patients was increased 8–9-fold compared to saline-infused patients, $F(1,23) = 86.70$, $p<0.0001$. Plasma-free tryptophan increased 10–12-fold, $F(1,24) = 47.10$, $p<0.0001$. This large difference between tryptophan-infused and control subjects was maintained throughout the experiment. At the beginning of the experiment plasma total tryptophan in the control subjects was below the range for normal healthy subjects (9000–15000 ng/ml) and four hours later the mean value was less than half the mean value for normal subjects. As can be seen in Table 1 the levels of other large neutral amino acids were also reduced though the reduction was not as great as for tryptophan. There were no differences between the tryptophan and saline-treated groups in the levels of the other large neutral amino acids.

Patients were given as much morphine as they required to feel comfortable. In spite of the dramatic effect of tryptophan loading on plasma tryptophan levels, the treatment did not reduce the amount of morphine required to control pain [for cumulative dose, $t(24) = 0.423$, NS] (Fig. 2), or reduce the patient's ratings of pain when they recovered from anesthesia, $t(24) = 1.361$, NS (Fig. 3).

TABLE 1

MEAN (STANDARD DEVIATION) PLASMA CONCENTRATIONS (μmol/l) OF LARGE NEUTRAL AMINO ACIDS IN PATIENTS INFUSED WITH TRYPTOPHAN OR SALINE, AND UNOPERATED SUBJECTS

	Saline	Tryptophan	Normal*
Threonine	107.0 (55.0)	95.0 (36.9)	171 (27)
Valine	181.0 (22.6)	187.1 (31.1)	247 (39)
Isoleucine	45.7 (6.8)	50.4 (18.4)	84 (14)
Leucine	95.4 (15.3)	100.6 (29.3)	156 (27)
Methionine	17.3 (5.4)	18.8 (8.3)	30 (4)
Tyrosine	36.6 (5.0)	42.1 (14.8)	60 (12)
Phenylalanine	37.7 (6.6)	40.2 (9.5)	72 (11)
Histidine	66.5 (24.9)	69.2 (20.5)	97 (15)
Tryptophan	30.7 (3.8)	256.5 (74.0)	64 (8)

*Approximate normal value in overnight fasted subjects from (46).

Indeed, the pain ratings were nonsignificantly higher for the tryptophan patients. It can be seen in Fig. 2 that, in both groups, patients required 12–14 mg in the first 30 min after admission to the recovery room. By 60 min, morphine requests had fallen to 4–5 mg and continued to decline slowly through the following two hours (see Fig. 2). Mean plasma morphine levels, taken during the third hour, were very similar for the two groups (53.1 ± 9.6 and 49.9 ± 11.9 ng/ml for saline and tryptophan groups, respectively).

Patients required an average of 25.1 mg ($n = 26$) of morphine to control pain during the first 3 hr in the recovery room, in addition to 11.2 mg they received during the operation. In spite of these relatively large doses, there were no untoward reactions to analgesic medication. During the hour between entry to the recovery room and establishing pain control, the breathing rate fell by approximately 2 breaths per minute and thereafter remained stable. There was no change in blood gases at this time and all vital signs remained within the normal range for the duration of the study period. There was no delayed effect of the tryptophan infusion. Tryptophan-infused patients received slightly more analgesic medication (codeine plus meperidine) over the three days following surgery, but the difference was not significant, $t(23) = 0.622$, NS.

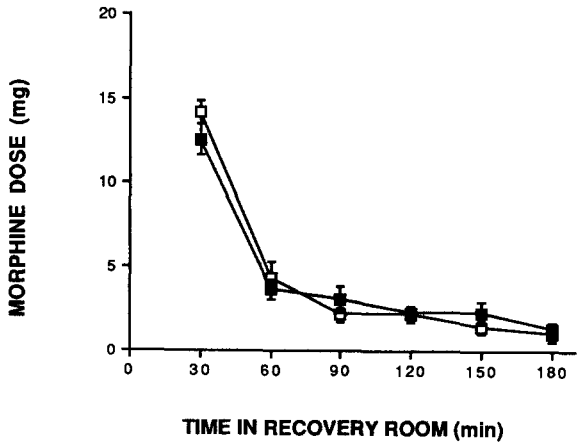

FIG. 2. The dose of morphine taken per 30 min during the first 180 min in the recovery room for patients infused with tryptophan (filled symbols) or saline (open symbols). Vertical bars are standard errors.

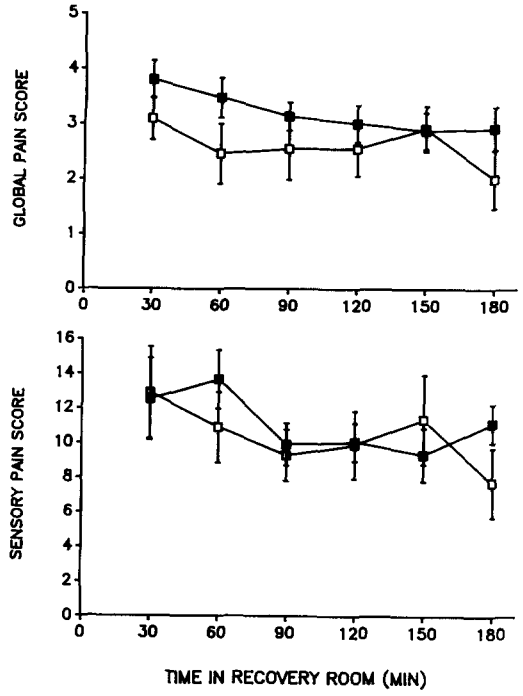

FIG. 3. Global pain scores (panel A) and Sensory pain scores (panel B) during the first 180 min in the recovery room for patients infused with tryptophan (filled squares) or saline (open squares). Vertical bars are standard errors.

Despite the absence of any significant intergroup differences, consideration of intragroup relationships indicates that tryptophan levels may have influenced morphine analgesia. Since tryptophan hydroxylase is approximately half saturated at normal levels of tryptophan, and plasma tryptophan levels were raised 5- to 10-fold higher than normal by tryptophan loading, tryptophan hydroxylase should have been close to saturation in all patients given the tryptophan infusion. Thus, in this group, there should be very little relationship between plasma tryptophan and brain 5-HT synthesis. In contrast, plasma tryptophan levels in the saline-treated group vary in the range below half-saturation and there should be a direct relationship between plasma tryptophan level and 5-HT synthesis. Thus, to the extent that morphine analgesia is related to 5-HT, there should be a relationship between tryptophan level and morphine requirement for the saline controls, but not for the tryptophan-loaded group.

The matrix of correlations between the biochemical and physical data on the patients (Table 2) showed that there was a complex relationship between plasma tryptophan and the morphine requirement. In the saline group the total morphine dose was positively related to plasma total tryptophan ($r = .762$, $p<0.01$), but in the tryptophan-treated group a weak negative correlation was seen. The correlations between free tryptophan and morphine dose showed a similar pattern, but were smaller. However, other variables were correlated with either tryptophan level or the morphine requirement. For both groups, body weight was negatively correlated with both morphine requirement and total plasma tryptophan level, while, in the saline group, the amount of time spent in the operating room was positively correlated with weight.

The relationships with body weight might be expected since the effective dose of any substance may be related to body weight. Moreover, it is known that surgery is more difficult, and presumably takes longer, with patients that have large amounts of body fat. Though none of the patients in this study were obese, there were several patients over 80 kg in the saline group. In the present context, the amount of surgical stress might be expected to correlate with pain and, thus, influence the morphine requirement. This interpretation was supported by the fact that time in the operating room was strongly correlated with amount of analgesics (codeine plus meperidine) administered over the first three postoperative days [for all 26 subjects, $r(25) = .688$, $p<0.001$]. These analgesics were given after the tryptophan levels would have returned to normal and were administered by the ward staff who were not involved in the experiment. Thus, the time in the operating room does seem to reflect the amount of surgical trauma.

To determine whether the correlation between plasma tryptophan and morphine requirement was due to the fact that both were correlated with other variables, we used multiple regression to explore the contributions of plasma tryptophan, body weight, and time in the operating room as predictors of the morphine requirement. Table 3 summarizes the results of tests of several models. For the saline controls a multiple regression model with the three predictors (time in OR, weight and mean total tryptophan) produced a highly significant multiple correlation [$r = .912$; $F(3,7) = 11.507$, $p<0.005$]. When the proportion of variance explained (r^2) was adjusted for the number of variables in the equation, the model accounted for 75.9% of the variance in morphine dose. From Table 3 it can be seen that when variables were withdrawn from the model one at a time, only tryptophan level produced a significant change in the predictability of the morphine requirement. The model containing weight and time in the operating room, as well as tryptophan, is a considerably better predictor than the reduced models. Figure 4 depicts an analysis of

TABLE 2

MATRIX OF CORRELATIONS BETWEEN POTENTIAL PREDICTORS (TOTAL TRYPTOPHAN, FREE TRYPTOPHAN, BODY WEIGHT AND TIME IN OPERATING ROOM) OF MORPHINE REQUIREMENT (DOSE) AND ANALGESICS USED ON THE WARD IN THE THREE DAYS FOLLOWING SURGERY (TOTANALG)

	Dose	TOTTRYP	FREETRYP	Weight	Time OR	TOTANALG
Dose	*	.763	.459	−.757	−.430	−.338
TOTTRYP	−.272	*	.662	−.489	.070	.067
Free TRYP	−.067	.909	*	−.175	.174	−.013
Weight	−.429	−.473	−.505	*	.606	.253
Time OR	.488	−.249	−.134	.052	*	.595
TOTANALG	.236	−.211	−.061	−.114	.739	*

Saline group correlations above the diagonal, tryptophan group below the diagonal.

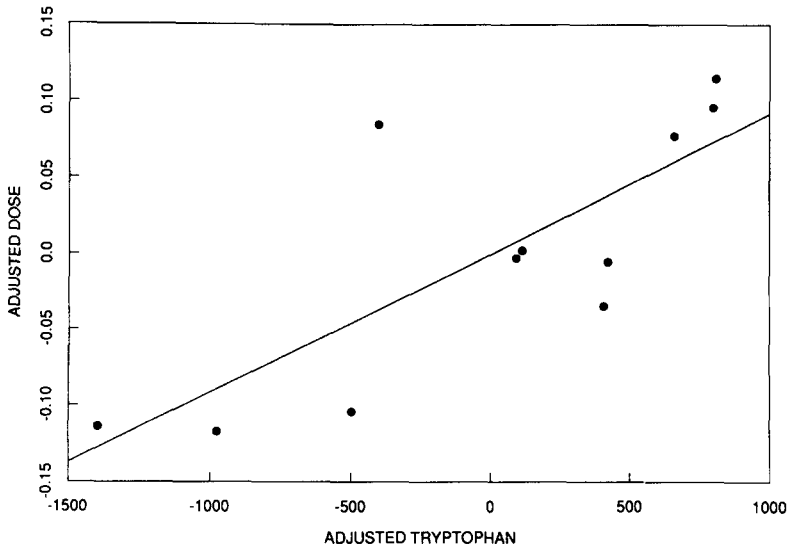

FIG. 4. Relationship between plasma total tryptophan concentration and the total dose of morphine received by patients infused with saline during 3 hr in the recovery room. Morphine dose and plasma tryptophan are adjusted for body weight and time in the operating room and a least squares regression line (r = .779) is fitted to these residuals. Graph generated by S-PLUS (Statistical Sciences Inc.).

residuals showing that, when weight and time in the OR are taken into account, plasma tryptophan level accounts for 60% of the variance in morphine requirement in the saline-infused group ($r^2 = .607$, $p<0.01$).

The lower panel of Table 3 shows the multiple regression analysis for the patients in the tryptophan group. Again, the model containing the three variables was a moderately good predictor of the morphine requirement (adjusted $r^2 = .522$), but the combination of weight and time in OR accounted for more than 50% of the variance. In this model, body weight was the major explanatory factor. Table 3 also shows that inclusion of tryptophan in the model has no significant impact on the amount of the variance explained.

TABLE 3

TESTS OF HYPOTHESES (SPSS-X REGRESSION) ON THE ROLE OF PLASMA TOTAL TRYPTOPHAN, BODY WEIGHT AND TIME IN THE OPERATING ROOM AS PREDICTORS OF THE DOSE OF MORPHINE REQUIRED TO CONTROL PAIN IN PATIENTS INFUSED WITH SALINE OR TRYPTOPHAN SOLUTION

Source of Variance	*Change in r^2	F	p
Saline Group			
Tryptophan	.258	10.708	0.014
Weight	.015	0.621	0.457
Time in OR	.057	2.352	0.169
Weight, time in OR	.250	5.186	0.042
Weight, time in OR, tryptophan	.831	11.507	0.004
Tryptophan Group			
Tryptophan	.179	5.256	0.043
Weight	.362	10.610	0.008
Time in OR	.149	4.368	0.061
Weight, time in OR	.550	8.058	0.007
Weight, time in OR, tryptophan	.624	6.094	0.011

*Difference in r^2 between the model with and without the variable(s).

Figures 2 and 3 show the time course of pain control measured three different ways—by morphine requirements, by global pain scores and sensory pain scores. Of the 17 sensory pain descriptors, only 8 (sharp-stabbing; burning; tight-squeezing; pressing; aching; cramping; penetrating; tender-sore) were given ratings of 2 or more by at least 25% of the patients. To compute the sensory pain score, the ratings of these 8 descriptors were summed for each patient. The time course of morphine requirements indicated that pain was satisfactorily controlled within the first hour in the recovery room. It may be of interest to note that rather large doses of morphine were necessary to make the patients comfortable. Clinical observation confirmed that pain control was achieved. When patients entered the recovery room they were distressed and uncommunicative, but as requests for morphine tapered off they became talkative or sleepy, and began to be concerned with incidental matters, such as their appearance. In contrast to these obvious signs of relief, the rating scales of pain intensity showed little change. There was no difference between saline- and tryptophan-infused patients in this regard. Tryptophan-infused patients had higher global pain scores when they entered the recovery room, though the difference was not significant. In fact, over the three hours of treatment in the recovery room, there was no significant reduction in global pain scores, $F(5,120) = 1.51$, NS, or in scores that measured specifically sensory aspects of pain (burning, tearing etc.), $F(5,120) = 1.53$, NS. This appears to reflect the often quoted phenomenon that, in the doses used clinically, opiates relieve suffering rather than block nociceptive inputs. Again, incidental notes support this interpretation. One patient was asked by the nurse if she was in pain and needed more morphine. The patient replied "No, I don't have pain—it hurts."

DISCUSSION

Our results indicate that tryptophan loading does not potentiate the analgesic action of morphine on postsurgical pain. On the contrary, when plasma tryptophan concentration is at or below the normal range, the level of plasma tryptophan appears to be

positively related to the amount of morphine required to control pain. The correlation was greater using total plasma tryptophan than using the free (i.e., nonalbumin bound) level. This is not surprising since the apparent free fraction in vivo can extend to either extreme (i.e., the total or free values measured in vitro) depending on the exact physiological circumstances (26).

The fact that tryptophan loading abolishes the relationship between plasma tryptophan and morphine requirement supports the hypothesis that the relationship is causal. The tryptophan infusion increased tryptophan concentrations by almost an order of magnitude so that, in these patients, tryptophan hydroxylase should have been fully saturated with tryptophan (40) and variability in the rate of 5-HT synthesis should have been reduced. The weak negative correlation between plasma tryptophan and morphine dose in tryptophan-infused subjects was probably not due to altered 5-HT synthesis, but to the fact that both variables were inversely related to body weight in this group. Plasma amino acid levels are reduced after surgery (12, 19). Because of competition between large neutral amino acids (LNAA) for transport across the blood-brain barrier, the availability of tryptophan to the brain is determined by the ratio of the concentration of tryptophan to the sum of the other LNAA (valine, isoleucine, phenylalanine, tyrosine and leucine) (25,27). In the control patients plasma tryptophan was reduced more than the levels of competing amino acids and the TRP/LNAA ratio was 0.08 compared to a normal ratio of 0.10 to 0.13 (5,37). Thus, in the saline-treated group, unlike in the tryptophan-treated group, CNS 5-HT may have varied quite widely between subjects because of the wide variation in plasma tryptophan levels and the lack of saturation of tryptophan hydroxylase. The strong positive relationship between morphine dose and plasma tryptophan in the saline-treated subjects, and the lack of such a relationship in the tryptophan treated patients, is, therefore, consistent with what is known about the relationship between tryptophan availability and 5-HT synthesis. The implication of this is that lowered 5-HT potentiates morphine analgesia. If this is so, then why was there no significant difference between the saline- and tryptophan-treated groups in morphine requirement and pain measures? One explanation is that the relationship between tryptophan levels and morphine requirement in the control group was not causal, but occurred only because both plasma tryptophan and morphine dose covaried with some other unknown factor. Alternatively, 5-HT levels may have to be reduced below their normal physiological levels for altered 5-HT to influence morphine analgesia. If this is so, the morphine requirements for those patients in the control group who had higher tryptophan would be in the same range as those seen in the tryptophan-treated group. In such circumstances one would expect to see a nonsignificant increase in pain scores or morphine requirements in the tryptophan group. This is what we did find (Fig. 4) and, we feel, on balance, that lowered 5-HT levels do potentiate morphine analgesia. This idea is supported by animal data as discussed below.

The clinical findings, described in the introduction, in which tryptophan had an analgesic effect, are consistent with the predictions derived from animal pain models based on reflex withdrawal from noxious stimuli (6, 7, 33). On the other hand, our present clinical findings parallel our results obtained with the formalin test model of injury-produced pain. In this test a subcutaneous injection of dilute formalin in an animal's paw produces inflammation and behavioural signs of discomfort for 1–2 hr (11). Decreases of 5-HT function increase the analgesic effect of morphine in the formalin test (2–4), while increased 5-HT function reduces analgesia (4,10). The present results thus support the view that there may be fundamental differences in the neurochemical substrates of morphine's effects on nocifensive reflexes and behaviours evoked by perceived pain and suffering, and suggests that the formalin test may be a valid model of the perceived pain and suffering which occurs postoperatively. Thus, while increased 5-HT function in the spinal cord may decrease nocifensive reflexes, increased brain 5-HT function may antagonize the clinical effect that morphine has on the response to perceived pain.

The fact that tryptophan did not reduce pain scores in our study suggests that tryptophan did not potentiate spinal 5-HT function sufficiently to diminish nociceptive inputs. Laboratory and clinical results indicate that morphine, in the doses used clinically, has relatively little effect on perceived pain intensity (15,32). In the present study, rather large doses (up to 32 mg in 4 hr) were administered with very good relief of suffering. Yet, even after several hours of pain control, the patient's ratings of the sensory intensity and quality of pain were not significantly reduced. It is unlikely that this finding is due to insensitivity of the numerical pain intensity scales used to assess pain. The scales used in this study do reflect the resolution of pain during the course of recovery from surgery (1). Moreover, verbal pain intensity, visual analogue and verbal pain relief scales are strongly correlated and show only minimal differences in sensitivity (18). A patient's refusal of available analgesic medication represents an objective behavioural criterion of pain relief, and, by this criterion, our patients rated pain relief as virtually complete. Our results, therefore, strongly support the idea that clinical analgesia produced by parenteral morphine represents a relief of suffering rather than blockade of nociceptive input (8) and suggests that the mechanisms involve the forebrain rather than the spinal cord. The challenge for the future will be to determine under which circumstances tryptophan will potentiate spinal 5-HT function to produce a clinically useful analgesia, and what controls the effect of tryptophan on the 5-HT system in brain that seems to antagonize morphine analgesia. The answers to these questions may lead to more specific analgesic treatments which are tailored to specific clinical circumstances.

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REFERENCES

1. Abbott, F. V.; Gray-Donald, K.; Johnston, C. C.; Edgar, L.; Jeans, M. E. Pain prevalence: A three week follow-up of hospitalized patients. Canadian and American Pain Societies Joint Meeting, Toronto, Canada; 1988 (abstract).
2. Abbott, F. V.; Melzack, R. Brainstem lesions dissociate neural mechanisms of morphine analgesia in different kinds of pain. Brain Res. 251:149–155; 1982.
3. Abbott, F. V.; Melzack, R.; Samuel, C. Morphine analgesia in the tail-flick and formalin pain tests is mediated by different neural systems. Exp. Neurol. 75:644–651; 1982.
4. Abbott, F. V.; Young, S. N. Effect of 5-hydroxytryptamine precursors on morphine analgesia in the formalin test. Pharmacol. Biochem. Behav. 31:855–860; 1988.
5. Ashley, D. V.; Barclay, D. V.; Chauffard, F. A.; Moennoz, D.; Leathwood, P. D. Plasma amino acid responses in humans to evening meals of differing nutritional composition. Am. J. Clin. Nutr. 36:143–153; 1982.
6. Basbaum, A. I.; Fields, H. L. Endogenous pain control mechanisms:

brainstem pathways and endorphin circuitry. Annu. Rev. Neurosci. 7:309–338; 1984.
7. Basbaum, A. I.; Fields, H. L. Endogenous pain control mechanisms: review and hypothesis. Ann. Neurol. 4:451–462; 1978.
8. Beecher, H. K. Measurement of subjective responses: Quantitative effects of drugs. New York: Oxford University Press; 1959.
9. Denckla, W. D.; Dewey, H. K. The determination of tryptophan in plasma, liver and urine. J. Lab. Clin. Med. 69:160–169; 1967.
10. Dennis, S. G.; Melzack, R. Pain modulation by 5-hydroxytryptaminergic agents and morphine as measured by three pain tests. Exp. Neurol. 69:260–270; 1980.
11. Dubuisson, D.; Dennis, S. G. The formalin test: A quantitative study of the analgesic effects of morphine, meperidine and brain stem stimulation in rats and cats. Pain 4:161–174; 1977.
12. Everson, T. C.; Fritschel, M. J. The effect of surgery on the plasma levels of the individual essential amino acids. Surgery 31:226–232; 1952.
13. Fernstrom, J. D.; Wurtman, R. J. Brain serotonin content: Physiological dependence on plasma tryptophan levels. Science 173:149–152; 1971.
14. Hosobuchi, Y.; Lamb, S.; Bascom, D. Tryptophan loading may reverse tolerance to opiate analgesics in humans: A preliminary report. Pain 9:161–169; 1980.
15. Keats, A. S.; Beecher, H. K. Pain relief with hypnotic doses of barbiturates and a hypothesis. J. Pharmacol. Exp. Ther. 100:1–13; 1950.
16. King, R. B. Pain and tryptophan. J. Neurosurg. 53:44–52; 1980.
17. Lieberman, H. R.; Corkin, S.; Spring, B. J.; Gordon, J. H.; Wurtman, R. J. Mood, performance, and pain sensitivity: changes induced by food constituents. J. Psychiatry Res. 17:135–145; 1983.
18. Littman, G. S.; Walker, B. R.; Schneider, B. E. Reassessment of verbal and visual analog ratings in analgesic studies. Clin. Pharmacol. Ther. 38:16–23; 1985.
19. Lund, J.; Sandin, R.; Sjernstrom, H.; Jorfeldt, L.; Wiklund, L. The influence of epidural analgesia on the splanchnic exchange of amino acids during upper abdominal surgery. Acta Anaesth. Scand. 31: 168–173; 1987.
20. Melzack, R. The McGill Pain Questionnaire: Major properties and scoring methods. Pain 1:277–299; 1975.
21. Messing, R. B.; Lytle, L. D. Serotonin-containing neurons: their possible role in pain and analgesia. Pain 4:1–21; 1977.
22. Mitchell, M. J.; Daines, G. E.; Thomas, B. L. Effect of L-tryptophan and phenylalanine on burning pain threshold. Physical Ther. 67: 203–205; 1987.
23. Moldofsky, H.; Lue, F. A. The relationship of alpha and delta EEG frequencies to pain and mood in 'fibrocitis' patients treated with chlorpromazine and L-tryptophan. EEG. Clin. Neurophysiol. 50: 71–80; 1980.
24. Nurmikko, T.; Pertovaara, A.; Pontinen, P. J.; Marnela, K. M.; Oja, S. S. Effect of L-tryptophan and supplementation on ischemic pain. Acupunct. Electrother. Res. 9:44–55; 1984.
25. Oldendorf, W. H.; Szabo, J. Amino acid assignment to one of three blood-brain barrier amino acid carriers. Am. J. Physiol. 230:94–98; 1976.
26. Pardridge, W. M. Tryptophan transport through the blood-brain barrier: in vivo measurement of free and albumin-bound amino acid. Life Sci. 25:1519–1528; 1979.
27. Pérez-Cruet, J.; Chase, T. N.; Murphy, D. L. Dietary regulation of brain tryptophan metabolism by plasma ratio of free tryptophan and neutral amino acids in humans. Nature 248:693–695; 1974.
28. Samanin, R.; Ghezzi, D.; Mauron, C.; Valzelli, L. Effect of midbrain raphe lesion on the antinociceptive action of morphine and other analgesics in rats. Psychopharmacologia 33:365–368; 1973.
29. Seltzer, S.; Dewart, D.; Pollack, R. L.; Jackson, E. J. The effects of dietary tryptophan on chronic maxillofacial pain and experimental pain tolerance. J. Psychiatry Res. 17:181–185; 1983.
30. Seltzer, S.; Stoch, R.; Marcus, R.; Jackson, E. J. Alteration of human pain thresholds by nutritional manipulation and L-tryptophan supplementation. Pain 13:385–392; 1982.
31. Shpeen, S. E.; Morse, D. R.; Furst, M. L. The effect of tryptophan on postoperative endodontic pain. Oral Surg. 58:446–449; 1984.
32. Smith, G. M.; Lowenstein, E.; Hubbard, J. H.; Beecher, H. K. Experimental pain produced by the submaximum effort tourniquet technique: further evidence of validity. J. Pharmacol. Exp. Ther. 163:468–474; 1968.
33. Takemori, A. E. Pharmacologic factors which alter the action of narcotic analgesics and antagonists. Ann. NY Acad. Sci. 281: 262–272; 1976.
34. Tenen, S. S. Antagonism of the analgesic effect of morphine and other drugs by p-chlorophenylalanine, a serotonin depletor. Psychopharmacologia 12:278–285; 1968.
35. Torgerson, W. S.; BenDebba, M. The structure of pain descriptors. In: Melzack, R., ed. Pain measurement and assessment. New York: Raven Press; 1983:49–54.
36. Vogt, M. The effect of lowering the 5-hydroxytryptamine content of the rat spinal cord on analgesia produced by morphine. J. Physiol. 236:483–498; 1974.
37. Weller, L. A.; Mergen, S.; Calloway, D. H.; Meissner, E. F. Serum amino acids in young men consuming diets differing in level and pattern of amino acids. Am. J. Clin. Nutr. 26:722–727; 1973.
38. Wurtman, R. J.; Hefti, F.; Melamed, E. Precursor control of neurotransmitter synthesis. Pharmacol. Rev. 32:315–335; 1981.
39. Yaksh, T. L. Direct evidence that spinal serotonin and noradrenaline terminals mediate the spinal antinociceptive effects of morphine in the periaqueductal gray. Brain Res. 160:180–185; 1979.
40. Young, S. N.; Gauthier, S. Effect of tryptophan administration on tryptophan, 5-hydroxyindoleacetic acid and indoleacetic acid in human lumbar and cisternal cerebrospinal fluid. J. Neurol. Neurosurg. Psychiatry 44:323–328; 1981.