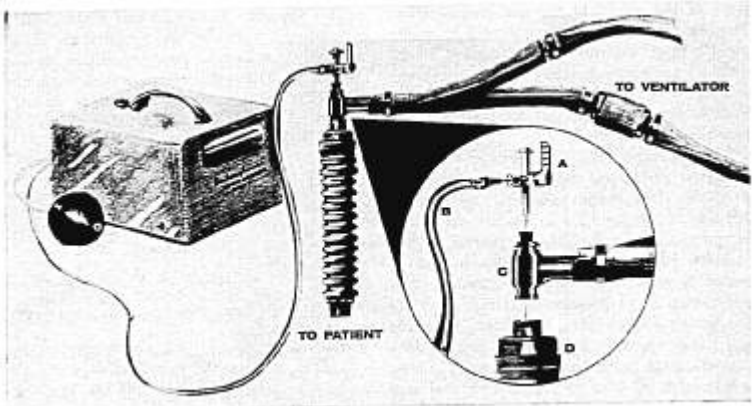

FIG. 1. Device for continuous oxygen measurement.

placed as shown in the figure the inspired partial pressure of oxygen can be monitored continuously as the gas drive of the ventilator will continuously fill the input line of the oxygen analyzer.

Adjustment of the control lever of the stopcock will also permit sampling of gas for measurement in electrode systems, continuous sampling for other measurements (*e.g.*, pneumotachygraph, CO_2 analyzer, etc.), cessation of flow through the O_2 analyzer, or cessation of flow through any portal of the stopcock. This device is inexpensive, simple to install or replace, and allows expansion of monitoring capabilities in patients who require constant ventilatory support.

Bupivacaine Hydrochloride: Laboratory and Clinical Studies

DANIEL C. MOORE, M.D., L. DONALD BRIDENBAUGH, M.D., PHILLIP O. BRIDENBAUGH, M.D., GEOFFREY T. TUCKER, PH.D.

A long-acting anilide local anesthetic agent, bupivacaine (1-n-butyl-DL-piperidine-2-carboxylic acid-2,6-dimethylanilide hydrochloride) (LAC 43, Marcaine), was synthesized in 1957 by Af Ekenstam *et al.*[1] Its chemical and pharmacologic properties have been studied in the laboratory, and at least 24 reports of its clinical use in Europe, Japan, and South America have appeared.[2-26]

The present study of bupivacaine was designed to: 1) measure bupivacaine in whole blood over a four-hour period following a single dose, as an index of elimination from the blood; 2) discern toxicity, *i.e.*, changes in blood morphology, blood chemistry; and urine; 3) correlate dosage with blood levels and de-

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velopment of complications; 4) confirm findings of others; and 5) meet the initial requirement of the Federal Food and Drug Administration for the introduction of an agent for clinical trial.

Methodology

The subjects of the study were 30 patients from whom informed consent had been obtained. Bupivacaine, 0.5 per cent, was administered to 15 patients with 1:200,000 epinephrine and to the other 15 without the vasoconstrictor. The maximum allowable dose of bupivacaine was 150 mg (30 ml).

Clinical Laboratory Tests: Prior to injection of the drug the following routine tests were done: 1) blood studies, including determination of hematocrit, hemoglobin, erythrocyte count, leukocyte count and differential; 2) prothrombin time, serum glutamic oxalacetic transaminase, serum alkaline phosphatase, blood urea nitrogen, and methemoglobin; 3) urinanalysis for specific gravity, sugar, *p*H, and sediment. These procedures were repeated on the second or third day after anesthesia and operation. Methemoglobulin determinations were also obtained at one- and four-hour intervals following injection of bupivacaine.

Blood Levels of Bupivacaine: A teflon needle was placed in an antecubital vein and a control blood sample was drawn. After regional block with bupivacaine, additional blood samples were drawn at approximately five, 20, 35, 45, 55, 65, 125, 185, and 245 minutes. Blood levels of bupivacaine in the 30 patients were determined by gas chromatography at one laboratory, and duplicate samples from eight cases were sent to another laboratory for the same determinations. The two laboratories used similar methods for the analyses.

The gas chromatographic studies reported herein were done prior to the establishment of our own method. That technique has since been modified * so that the standard deviation of ±8 per cent at 1.0 μg/ml levels or above has been reduced to ±4 per cent at 0.04 μg/ml levels or above. Using this technique, peak levels in whole blood following the injection of 150 mg of bupivacaine into the epidural space have ranged from 0.4 to 1.0 μg/ml, with a mean of 0.6 μg/ml, occurring in 15 to 20 minutes.

Determination of Onset and Sensory Levels of Anesthesia: Allis forceps were used to determine absence of sensation. When hypesthesia to partial closure (not to first ratchet) of the forceps occurred this was recorded as the time of onset. When the patient felt no pain with the Allis forceps closed to the first ratchet, and when the anesthetic level did not rise above that dermatome, the time was noted.

Results

Every patient had a satisfactory block, that is, one in which sensory and motor anesthesia was adequate for the surgical procedure. Of the 30 patients, 21 were women and nine, men, ranging from 20 to 75 years in age. Epidural block was administered to 21 patients, caudal block to six, sciatic and femoral block to two, and brachial plexus block to one. In

* The modified assay procedure for bupivacaine is as follows:

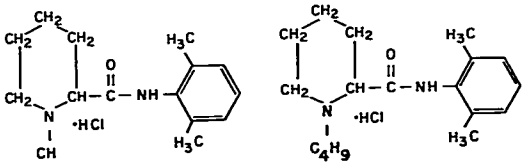
Blood and plasma: Mix 1 ml whole blood or 1/2 ml plasma + 1 ml internal marker solution (cyclizine HCl) 0.4 μg/ml + 0.2 ml 0.5 N NaOH. Shake 1 min with 6 ml freshly distilled ether; centrifuge, freeze in dry ice/acetone bath; decant off ether. Ether extract back-extracted into 1 ml 0.1 N HCl; shake 1 min; centrifuge; freeze, decant and discard ether. Add 0.5 ml 0.5 N NaOH to acidic solution. Extract with 6 ml ether as before. Collect ether in evaporating tube with tapered base. Add small boiling chip and evaporate at 43 C just to dryness. Take up residue in 5 μl carbon disulfide and inject into gas chromatograph.

Urine: Mix 1 to 5 ml urine + 1 ml internal marker solution (cyclizine HCl) + 0.5 ml 0.5 N NaOH. Extract with 2 × 2.5 ml ether (5 min on tilt shaker); centrifuge; freeze; collect ether in evaporating tube. Evaporate to about 20 μl. Inject 1 to 2 μl into gas chromatograph.

Gas chromatographic conditions: Varian series 2100 gas chromatograph; column—3 per cent OV-17 on Gas-Chrom Q, 100–120 mesh (A/W, DMCS treated); glass 6′ × ¼″ i.d. tubing. Oven 235 C; injector 260 C, detector (F.I.D.) 260 C, N_2 30 ml/min.

Retention times: cyclizine, 4.5 min; pipecholylxylidine, 6.0 min; bupivacaine, 9.9 min. (Pipecholylxylidine is a metabolite of bupivacaine. For quantitative analysis this is chromatgraphed as its acetyl derivative, retention time 13.4 min, prepared by adding 1 μl acetic anhydride to final ether extract prior to concentration.)

All peaks are sharp and symmetrical. Measure peak height ratios (drug or metabolite to internal marker). *N.B.:* In whole blood only, large peaks appearing after assay of six samples preclude further use of column for about 80 min. Overcome delay by using a dual-column instrument.



Mepivacaine Bupivacaine

FIG. 1. Chemical structures of mepivacaine and bupivacaine.

the 29 patients who underwent surgery, the 33 operations performed were: appendectomy 1, bunionectomy 1, cholecystectomy 1, gastric resection 1, hemorrhoidectomy 3, hiatus hernia repair 1, hysterectomy 8, inguinal hernia 2, Marshall-Marchetti operation 4, nephrectomy 1, pilonidal cystectomy 1, perineal prostatectomy 1, perineorrhaphy 3, tendon (Achilles) repair 1, tendon and nerve repair of forearm 1, transurethral resection 1, operation for varicose veins 1, repair of vesicovaginal fistula 1.

Dosage, Onset, Establishment of Anesthesia, and Duration: Bupivacaine was administered in doses ranging from 11 ml (55 mg) to 30 ml (150 mg). Onset of analgesia occurred in four to seven minutes, with maximum surgical analgesia developing in seven to 25 minutes. Duration of sensory anesthesia varied with the level of anesthesia in epidural and caudal block. The skin of the upper abdomen (T4 to T10) was anesthetized for an average of three and a half hours; the skin of the lower abdomen (T10 through T12), four and a half hours; the extremities, six hours; the perineum, eight and a half hours. In the 15 patients who received bupivacaine with 1:200,000 epinephrine, the duration of anesthesia was not consistently prolonged. Supraclavicular brachial block in one instance lasted nine hours, and the two sciatic and femoral blocks lasted eight and a half and 17 hours, respectively.

Postoperative Laboratory Tests and Blood Levels: Significant elevations above the normal values for blood urea nitrogen, serum glutamic oxalacetic transaminase, and serum alkaline phosphatase occurred in one, nine, and four patients, respectively. Results of all other blood and urine tests were within normal ranges for the operative procedures performed.

Bupivacaine could be detected in the blood within five minutes. Peak blood levels of 0.14 to 1.18 μg/ml were found in from five minutes to two hours, and gradually declined to 0.1 to 0.34 μg/ml in four hours, depending upon the nature of the block.

Complications: There were no general systemic or local toxic reactions to the drug. In one patient, immediately following the injection of the drug and the turning of the patient from the prone position to the supine position, a trigeminal cardiac arrhythmia developed.

Ten patients developed marked peripheral vasoconstriction in the unanesthetized extremities. Of these, two received solutions of bupivacaine without epinephrine; in eight the solutions contained epinephrine.

DISCUSSION

Pharmacology: Bupivacaine differs from mepivacaine in that a butyl group is substituted for a methyl group (fig. 1). The following observations have been made in mice, guinea pigs, and rabbits: 1) bupivacaine, though chemically closely related to mepivacaine (Carbocaine), is more like tetracaine (Pontocaine) in local anesthetic and toxicologic properties; 2) the acute toxicity (LD_{50}) of bupivacaine is about the same as that of tetracaine, and approximately four times higher than that of mepivacaine; 3) bupivacaine and tetracaine have the same tissue toxicity, about six times higher than that of mepivacaine; 4) in regional block (local, epidural, and caudal block) bupivacaine and tetracaine are approximately three times more potent than mepivacaine.[6]

Blood Levels: Following a single dose of 150 mg of bupivacaine, in epidural, caudal, or peripheral nerve block, venous levels of approximately one μg/ml of whole blood or less may be expected. The blood levels of bupivacaine showed no significant relationship either to total dose per pound of body weight, or to addition to epinephrine to the solution.

The highest blood levels of 0.34 μg of bupivacaine/ml of blood, present 245 minutes after administration, indicate that the agent is not eliminated rapidly from the human body. Furthermore, since this initial study of single-dose injections of bupivacaine, determinations of blood levels of bupivacaine following refill doses in a continuous technique show that it does accumulate like the closely-related agent, mepivacaine.[27]

Toxicology: With the exception of those noted, the results of clinical laboratory tests done following surgery did not differ from the preoperative control values beyond what might be expected as a result of the surgical procedures (table 1). In patients not undergoing operation, but having only diagnostic or therapeutic blocks, Dhuner found no change in blood morphology.[10]

In one case in which the blood urea nitrogen was significantly elevated, the patient was found to have an advanced ovarian malignancy, which accounted for the elevation. Elevations of serum glutamic oxalacetic transaminase in nine patients resulted from tissue trauma from the operations.

In the opinion of the pathologist reviewing the laboratory results, the elevation of serum alkaline phosphatase in four patients could not be explained on the basis of operation alone (hysterectomy, repair of inguinal hernia, Marshall-Marchetti operation, perineal

TABLE 1. Results of Laboratory Studies of 30 Patients before and after Administration of Bupivacaine*

	Before Mean ± SD	After Mean ± SD	Normal
Hematology			
Hematocrit (per cent)	41.9 ± 4.3	38.6 ± 4.5	35–45
Hemoglobin (g/100 ml)	13.9 ± 1.5	12.7 ± 1.3	13.5–16
Erythrocyte count (millions)	4.61 ± 0.56	4.16 ± 0.51	3.8–5.5
Leukocyte count	7,761 ± 2,421	9,391 ± 2,793	5,000–10,000
Segmented cells	56.6 ± 11.4	68.9 ± 10.4	40–75
Band cells	2.6 ± 3.1	5.2 ± 7.0	<10
Lymphocytes	33.2 ± 10.8	19.0 ± 7.2	20–45
Monocytes	5.0 ± 3.0	4.6 ± 2.6	2–10
Eosinophils	2.0 ± 2.3	2.4 ± 2.0	1–6
Basophils	0.5 ± 1.1	0.13 ± 0.35	1
Blood chemistry			
Prothrombin time (sec)	14.3 ± 1.2	13.8 ± 1.0	12–15
SGOT (IU/l)	12.4 ± 4.3	20.6 ± 11.9	5–20
Serum alkaline phosphatase (IU/l)	48.3 ± 14.7	58.6 ± 29.7	25–90
BUN (mg/100 ml)	13.4 ± 3.6	11.5 ± 4.7	14–22
Methemoglobin (gm/100 ml)	—	—	0–0.7
Routine	0.22 ± 0.11	0.23 ± 0.10	
1 hour	—	0.22 ± 0.09	
4 hours	—	0.21 ± 0.06	
Urinalysis			
Specific gravity	1.013 ± 0.008	1.012 ± 0.007	1.001– 1.030
Sugar	0	0	0
pH	6.2 ± 0.7	6.3 ± 1.0	5–7
Sediment	—	Not unusual	—

* Standard deviation: One SD on either side of the mean includes 67 per cent of the population. Two SD on either side of the mean include 95 per cent of the population. Three SD on either side of the mean include 99 per cent of the population.

prostatectomy). Therefore, the question of derangement of hepatic metabolism was considered. In no patient did jaundice occur postoperatively, and all patients had uneventful postoperative anesthetic courses. These patients have been followed for a year and have reported no complications.

Clinical Properties: The qualities of bupivacaine noted by other investigators and confirmed by this study include: 1) prolonged duration of sensory anesthesia, which is probably its most outstanding characteristic: anesthesia lasts two to three times longer than with mepivacaine or lidocaine and 20 to 25 per cent longer than with tetracaine; 2) absence of nerve damage; 3) onset of anesthesia comparable to mepivacaine or lidocaine; 4) no greater occurrence of generalized systemic toxic reactions than when lidocaine or mepivacaine is used; and 5) lack of tissue toxicity. However, we found that following single-dose epidural block muscle relaxation during intra-abdominal procedures was not profound in approximately 25 per cent of the patients, compared with 1.5 per cent after mepivacaine or lidocaine. In all cases sensory anesthesia was excellent, comparable to that produced by mepivacaine or lidocaine.

Complications: The marked peripheral vasoconstriction which occurred in the unanesthetized area of the patient had the following characteristics: 1) it appeared approximately one and a half to two hours after injection of bupivacaine; 2) it lasted two to four hours; 3) it was perhaps somewhat dose-related; it was not observed in patients who had small doses (10 to 15 ml); 4) withdrawal of blood samples from the patients' antecubital veins was difficult. This phenomenon does not occur only with bupivacaine, although it is more prolonged than with lidocaine and mepivacaine. We are investigating this phenomenon, and have found that peripheral vasoconstriction in the unanesthetized area follows spinal, epidural and caudal blocks, as the body tries to compensate for vasodilatation in the blocked area.

The one patient who developed trigeminal rhythm had normal electrocardiograms in 1952, 1961, and 1963, but on November 6, 1967, extrasystoles were noted. On November 10, 1967, the patient was brought to surgery for a radical perineal prostatectomy. The blood pressure was 120/76 mm Hg, pulse rate was 54 beats/min. Plastic tubing was placed in the caudal canal and bupivacaine, 15 ml, with 1:200,000 epinephrine was injected. Within two minutes, the patient was turned supine, and blood pressure and pulse were checked immediately. Blood pressure was 140/60 mm Hg. Pulse rate was 96 beats/min and irregular. Anesthesia to the level of the tenth thoracic dermatome resulted. The electrocardiogram confirmed the diagnosis of trigeminal rhythm. The surgical operation was cancelled. Twenty hours after the onset of the irregularity, the rhythm reverted to normal.

Although no other complications were observed in the 30 patients during hospitalization, the following problems have been reported in a total of 1,726 patients by others: 1) convulsions from inadvertent intravenous doses in five patients; 2) tremors, shivering or fainting in 31; 3) bradycardia in 16; 4) nausea or vomiting, or both, in 28; 5) headache in 11; 6) paresthesias for three weeks in one; 7) inadvertent high spinal anesthesia to the second and fifth thoracic dermatome in two; 8) approximately 24 hours of anesthesia in one.[8, 10, 11, 17, 24, 25]

Conclusions

The long duration of action of bupivacaine makes it a desirable local anesthetic agent when the physician selects peripheral nerve block, epidural block, or caudal block for a long surgical or obstetrical procedure, relief of postoperative pain, or to produce prolonged vasodilatation of an extremity.

Further extensive study of bupivacaine must be undertaken to provide the following data: 1) uptake, distribution, and excretion in man following peripheral nerve block, caudal block, and epidural block; 2) levels of drug in the blood at which systemic toxic reactions are likely to occur, as well as the concentration and volume likely to produce such levels; 3) toxicity to specific organs, for example, the liver; 4) effects on the newborn; 5) extent of cumulation after repeated doses in continuous techniques; 6) duration of anesthesia compared with anesthesia produced by other local

anesthetics, using the patient as his own control; 7) complications which may not have been observed in the 30 patients in this study.

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References

1. Af Ekenstam, B., Egner, B., and Pettersson, G.: N-alkyl pyrolidine and n-alkyl piperidine carboxylic acid amides, Acta Chem. Scand. 11: 1183, 1957.
2. Telivuo, L.: A new long-acting local anaesthetic solution for pain relief after thoracotomy, Ann. Chir. Gynaec. Fenn. 52: 513, 1963.
3. Widman, B.: Clinical trial of a new local anaesthetic (LAC-43) with the aid of the pinprick and ninhydrin methods in finger blocks, Acta Anaesth. Scand. 8: 219, 1964.
4. Albert, J., and Lofstrom, B.: Bilateral ulnar nerve blocks for the evaluation of local anaesthetic agents, Acta Anaesth. Scand. 9: 1, 1905.
5. Lofstrom, B., Wennberg, A., and Widen, L.: Late disturbances in nerve function after block with local anaesthetic agents, Acta Anaesth. Scand. 10: 111, 1966.
6. Henn, F., and Brattsand, R.: Some pharmacological and toxicological properties of a new long-acting local analgesic, LAC-43 (Marcaine), in comparison with mepivacaine and tetracaine, Acta Anaesth. Scand. Suppl. XXI, 9, 1966.
7. Hollmen, A.: Axillary plexus block, Acta Anaesth. Scand. Suppl. XXI, 53, 1966.
8. Herbring, B. G.: A comparative study of LAC-43, mepivacaine and tetracaine in caudal anaesthesia, Acta Anaesth. Scand. Suppl. XXI, 45, 1966.
9. Ekblom, L., and Widman, B.: A comparison of the properties of LAC-43, prilocaine and mepivacaine in extradural anaesthesia, Acta Anaesth. Scand. Suppl. XXI, 10: 33, 1966.
10. Dhuner, K.-G.: Clinical experience with Marcaine (LAC-43), a new local anaesthetic, Acta Anaesth. Scand. Suppl. XXIII, 395, 1966.
11. Ekblom, L., and Widman, B.: LAC-43 and tetracaine in spinal anaesthesia, Acta Anaesth. Scand. Suppl. XXIII, 419, 1966.
12. Korkeila, J.: LAC-43—clinical experiences with a new long-acting local anaesthetic agent in epidural anaesthesia, Acta Anaesth. Scand. Suppl. XXIII, 453, 1966.
13. Castren, J. A., and Tammisto, T.: A clinical evaluation of a new local anaesthetic (Marcaine-adrenalin) in ocular surgery, Acta Ophthal. (Kobenhavn) 44: 837, 1966.
14. Bergh, N., Dottori, O., Axson Lof, B., Simonsson, B., and Ygge, H.: Effect of intercostal block on lung function after thoracotomy, Acta Anaesth. Scand. Suppl. XXIV, 85, 1966.
15. Nolte, H., Dudeck, J., Brade, R., and Bolch, H.: Marcain (LAC-43)—ein neues, langwirkendes lokalanaestheticum, Der Anaesthesist 7: 229, 1966.
16. Telivuo, L., and Perttala, Y.: Use of x-ray contrast medium to control intercostal nerve blocks, Ann. Chir. Gynaec. Fenn. 55: 185, 166.
17. Hollmen, A.: Experience with LAC-43, a new anaesthetic with prolonged action, in operative and therapeutic epidural and regional anaesthesia, Ann. Chir. Gynaec. Fenn. 55: 322, 1966.
18. Widman, B.: Some circulatory and respiratory effects of intravenously infused local anaesthetics, Acta Anaesth. Scand. Suppl. XXV, 34, 1966.
19. Af Ekenstam, B.: The effect of the structural variations on the local analgetic properties of the most commonly used groups of substances, Acta Anaesth. Scand. Suppl. XXV, 10, 1966.
20. Tammisto, T., and Sivula, A.: Schmerzbekampfung nach laparotomien mit dem neuen lokalanaestheticum Marcain, Der Anaesthesist 1: 1, 1967.
21. Telivuo, L.: Effects of intra-arterial mepivacaine and bupivacaine on neuromuscular transmission in man, Acta Anaesth. Scand. 11: 327, 1967.
22. Stockhausen, H.: Erfahrungen mit einem neuen lokalanaesthetikum bei der parazervikalblockade, Deutsch. Med. Wschr. 48: 2220, 1967.
23. Puente-Egido, J., Dudeck, J., and Nolte, H.: Die klinische anwendung des lokalanaestheticums Marcain (Carbostesin), Der Anaesthesist 6: 224, 1967.
24. Watt, M. J., Ross, D. M., and Atkinson, R. S.: A clinical trial of bupivacaine, Anaesthesia 23: 2, 1968.
25. Steel, G. C., and Massey Dawkins, C. J.: Extradural lumbar block with bupivacaine (Marcaine: LAC-43), Anaesthesia 23: 14, 1968.
26. Duthie, A. M., Wyman, J. B., and Lewis, G. A.: Bupivacaine in labor, Anesthesia 23: 20, 1968.
27. Moore, D. C., Bridenbaugh, L. D., Bagdi, P. A., and Bridenbaugh, P. O.: Accumulation of mepivacaine hydrochloride after caudal block, Anesthesiology 29: 585, 1968.