Original Research Article

Indian Journal of Anesthesia and Analgesia
2019; 6(5) (Part - I): 1547-1552
DOI: http://dx.doi.org/10.21088/ijaa.2349.8471.6519.9

Ketamine as an Adjunct with Bupivacaine in USG Guided Paravertebral Analgesia for Modified Radical Mastectomy

Rajeev Prajapat[1], Om Prakash Suthar[2], ML Tak[3]

[1]Consultant Anaesthesiologist, Department of Anesthesiology, Shri Sidhivinayak Hospital, Sumerpur, Pali, Rajasthan 306902, India. [2]Associate Professor, Department of Anesthesiology, Government Medical College, Pali, Rajasthan 306401, India. [3]Professor, Department of Anesthesiology and Critical Care, Dr. SN Medical College and Associated Group of Hospitals, Jodhpur, Rajasthan 342003, India.

Abstract

Background: Adjuvants like fentanyl and clonidine have found to prolong the duration of analgesia when used along with local anesthetic in the paravertebral space for breast surgery. This study was planned to study the effect of addition of ketamine to bupivacaine for paravertebral block on intra-operative and post-operative analgesia in patient undergoing modified radical mastectomy under general anesthesia. *Materials and Methods:* This prospective, randomized, controlled double blind study was conducted in 60 women of ASA grade I–III age between 18 to 70 years who underwent modified radical mastectomy. Group A consisted of 30 patients receiving PVB with 0.3 ml/kg of 0.25% bupivacaine and 1 ml normal saline prior to GA and Group B consisted of 30 patients receiving PVB with 0.5 mg/kg ketamine along with 0.3 ml/kg of 0.25% bupivacaine in normal saline prior to GA. Intra-operative supplemental fentanyl consumption, hemodynamic parameter, pain score and post-operative morphine consumption were compared. *Results:* The mean intra-operative fentanyl consumption requirement in group A was 21.95 ± 21.58 µg, and 12.83 ± 19.93 µg in group B. (p = 0.828) 60% of the patients in group A did not require any analgesic supplementation which was comparable to that in group B (63.33%). First requirement of rescue analgesia in post-operative period was after 3.63 ± 2.55 hr in group A and 3.13 ± 2.84 hr in group B, (p = 0.480). The mean VAS values in both the groups were statistically comparable at rest and as well as on movement. (p > 0.05). *Conclusion:* The present study, showed that the addition of ketamine to bupivacaine did not improve the efficacy or duration of paravertebral analgesia in the post-operative and intra-operative period in patients undergoing modified radical mastectomy.

Keywords: Paravertebral block; Analgesia; Ketamine; Bupivacaine; Modified radical mastectomy.

How to cite this article:

Rajeev Prajapat, Om Prakash Suthar, ML Tak. Ketamine as an Adjunct with Bupivacaine in USG Guided Paravertebral Analgesia for Modified Radical Mastectomy. Indian J Anesth Analg. 2019;6(5 Part-1):1547-1552.

Introduction

Surgery in form of either lumpectomy or modified radical mastectomy with axillary node dissection, in combination with chemotherapy or radiotherapy remains the treatment of choice for breast cancer. Modified Radical Mastectomy (MRM) includes removal of the entire breast and axillary dissection, in which levels I and II of axillary lymph nodes are removed. Breast surgery is frequently associated with nausea, vomiting, pain and pain restricted movement.

Pain, according to definition endorsed by the International Association for the Study of Pain

Corresponding Author: Om Prakash Suthar, Associate Professor, Department of Anesthesiology, Goverment Medical College, Pali, Rajasthan 306401, India.

E-mail: omeenipom12@gmail.com

Received on 11.05.2019, **Accepted on** 08.06.2019

(IASP), is "unpleasant sensory and emotional experience associated with actual or potential tissue damage, or described in term of such damage.[1] Post-operative pain managed in adequately, is documented to have several pathophysiological as well as economic implications, *e.g*., increased morbidity, duration of hospital stay and cost of medical care.

A wide variety of analgesic technique like local anesthetic infilteration, paravertebral and neuroxial analgesia, anti-convulsant, anti-neuropathic analgesic and NMDA (*N*-Methyl *D*-Aspartate) antagonist, apart from opioids based technique are employed for managing post-operative surgical pain following breast surgery. It is increasingly recognized that complex chronic pain syndrome may develop months to years later, if this acute post-operative pain is left untreated or undertreated.[2]

Paravertebral Block (PVB) for MRM was first described in April 1994 by Greegrass *et al*. He adapted the block for use in breast surgery, after surgeons asked for a way to prevent the intense side effects caused by general anesthesia (GA) precluding ambulatory surgery.[3] The technique used was modified by Eason and Wyatts' technique, which was simple to administered.[4] Before Eason and Wyatts described and standardized the technique, various other techniques were used by different authors.

PVB given prior to induction of GA, for breast surgery is known to provide improved intra-operative and post-operative analgesia, decreased incidence of nausea and vomiting, reduced surgical stress response and improved patient satisfaction. Therefore, Thoracic Paravertebral Block (TPVB) is the technique of injecting local anesthetic adjacent to the thoracic vertebra close to where the spinal nerve emerges from the intervertebral foramen. This result in ipsilateral somatic and sympathetic nerve blockade in multiple contiguous dermatomes above and below the injection site.[5]

Adjuvants like fentanyl and clonidine have already been used along with local anesthetic in the paravertebral space for breast surgery and have found to prolong the duration of analgesia.[6] The addition of ketamine to a local anesthetic or other analgesic in peripheral or neuroaxial anesthesia and analgesia improve or prolong pain relief.[7,8] Hence, this study was planned to study the effect of addition of ketamine to bupivacaine for paravertebral block on intra-operative and post-operative analgesia in patient undergoing modified radical mastectomy under general anesthesia.

Materials and Methods

This prospective, randomized, controlled double blind study was conducted in the Department of Anesthesiology and Intensive Care Unit at SN Medical College, Jodhpur. 60 women of ASA Grade I–III age between *18* and *70 years* who underwent modified radical mastectomy were included in study only after approval from Institution Ethical Committee and written informed consent from each patient were taken. Patients with local sepsis at site of block, severe chest wall deformity, coagulopathy or patient receiving any anticoagulants (platelet < 1,00,000), INR > 1.5, known hypersensitivity to amide type of local anesthetics, pregnancy, breast feeding and severe obesity BMI > *35 kg/m*^{2} were excluded from the study. The patients selected for the study were allocated to either group A or B using computer generated list of random permutations in double blind manner:

Group A: Consisted of patients receiving PVB with bupivacaine prior to GA. (*0.3 ml/kg* of 0.25% bupivacaine and *1 ml* normal saline).

Group B: Consisted of patients receiving PVB with bupivacaine with ketamine prior to GA (*0.5 mg/kg* ketamine along with *0.3 ml/kg* of 0.25% bupivacaine in normal saline).

The test solution was prepared by a fellow anesthesiologist who was not involved in the study so as to double blind the study. All the selected patients underwent a routine pre-anesthetic assessment, including explanation to the patient on post-operative pain assessment scale through visual analog scale (VAS). All the patients included in the study were premedicated with oral alprazolam *0.25 mg* two hours before the procedure. All of them were properly explained regarding the procedure of giving paravertebral block and were pre-loaded with *10–15 ml/kg* ringer lactate after I.V. line established with 18 G cannula on opposite hand. Baseline parameters including pulse rate, Non-invasive Blood Pressure (NIBP), oxygen saturation (SpO_2) and Respiratory Rate (RR) were recorded before starting PVB and before induction. The paravertebral block was performed prior to induction of general anesthesia.

All the patients received general anesthesia without testing the sensory level attained by TPVB. All patients premedicated with ondansetron *4 mg* and glycopyrrolate *0.2 mg*, then patients were induced with propofol (*1.5–2.5 mg/kg*, I.V.). Muscle relaxation was provided with vecuronium bromide after confirming adequacy of ventilation after loss of consciousness with propofol and the airway

was secured with appropriate size of endotraheal tube. The anesthesia was maintained with oxygen, isoflurane and vecuronium bromide. The $EtCO_2$ was maintained between *35–40 mm Hg*. Supplemental analgesia was provided with fentanyl *(0.5 µg/kg*) on the basis of rise in heart rate or systolic blood pressure by more than 20% of the base values for more than *5 minutes* (inadequate PVB was suspected). No other analgesia was administered intra-operatively. The number of doses and the total amount of supplement analgesia with fentanyl intra operatively was recorded for comparison between the two groups. Mephentermine *6 mg* was given in I.V. incremental dose to treat hypotension.

Continuous monitoring of HR, NIBP, SpO_2 and $EtCO_2$ were done in the intra-operative period and these were recorded every *15 minutes*. By end of surgery vecuronium effect was antagonized by I.V. Neostigmine *50 µg/kg* (max. *5 mg*) with glycopyrrolate *5 µg/kg*. After emergence patients were transferred to recovery room for a *2 hour* observation period. Analgesia in recovery room was provided by morphine *1 mg* I.V. as rescue medication, if needed, every *10 minutes* (a maxi. limit of *20 mg* in *4 hours*) until pain VAS score was ≤ 3. Time to first using the morphine was recorded and total dose of morphine was also calculated. HR, NIBP, SpO_2, RR and VAS score were also recorded at 0, 2, 6, 12 and 24 hours after the surgery. Ondansetron (*4 mg* I.V.) was given *8 hourly* as needed. Any psychomimetic changes (defined by agitation, hallucination or vivid dream) were also reported.

Data was recorded in 'Microsoft excel 2007' format and analyzed using SPSS version 15.0'. Continuous variable (age, weight, intra-operative supplemental fentanyl consumption, duration of surgery, hemodynamic parameter, respiratory rate, PONV scores, pain score and post- operative morphine consumption) were compared and analyze using student "*t*" test. Qualitative data (presence or absence of side effects and rescue anti-emetic drugs use) were analyzed using the chi-square test or Fischer's test, whichever applicable. A *p* - value less than 0.05 was considered significant for all parameters.

Results

Demographic data of all the patients in both the groups were found comparable ($p > 0.05$). The mean age of the patients in group A was *51.03 ± 13.81 years* compared to 54.4 ± 5.67 in group B. This difference was statistically not significant ($p = 0.757$) (**Table 1**).

The fentanyl consumption was found between *0 and 75 µg* in group A, *0 and 70 µg* in group B. The mean intra-operative fentanyl consumption requirement in group A was *21.95 ± 21.58 µg*, while in group B it was *12.83 ± 19.93 µg*. This difference was statistically non-significant.

($p = 0.828$) 60% of the patients in group A did not require any analgesic supplementation which was comparable to that in group B, where 63.33% of the patients did not require any supplementation of fentanyl (**Table 1**).

First requirement of rescue analgesia in post-operative period was after *3.63 ± 2.55 hr* and *3.13 ± 2.84 hr* in group A and B respectively which was statistically non-significant. ($p = 0.480$) 17 (28.33%) out of 60 patients consumed more than *30 mg* of morphine in *24 hours*. Among these 8 were in group A and 9 patients from group B. The cumulative consumption of morphine in both the group was comparable. ($p > 0.05$) (**Table 1**).

The mean VAS values in both the groups were statistically comparable at rest and as well as on movement (coughing) ($p > 0.05$) (**Table 2**).

Intra-operative and post-operative heart rate, systolic blood pressure and diastolic blood pressure, SpO_2 and $EtCO_2$ were recorded at *0, 2, 6, 12* and *24 hours* post-operatively. The recording at all interval were found comparable in both

Table 1: Demographic and other characteristics of both the groups

	Group A (Mean ± SD)	Group B (Mean ± SD)	*p* - value*
Age (*yrs*)	51.03 ± 13.81	49.93 ± 13.67	0.757
Weight (*kg*)	54.4 ± 5.67	54.53 ± 7.83	0.104
ASA grade	1.77 ± 0.49	1.83 ± 0.637	0.658
Mean duration of surgery (minutes)	103.83 ± 16.15	108.33 ± 14.47	0.293
Intra-operative fentanyl consumption (*ug*)	21.95 ± 21.58	12.83 ± 19.93	0.828
Time of first requirement of rescue analgesia-morphine (*hr*)	3.63 ± 2.55	3.13 ± 2.84	0.480
24 consumption of rescue analgesia-morphine (*mg*)	24.06 ± 7.239	25. 27 ± 5.965	0.486

*p - value (> 0.05) non-significant for all parameters.

Table 2: Post-operative VAS score at rest and on movement

Time of measurement (hr)	VAS score at rest (Mean ± SD)			VAS score on movement (Mean ± SD)		
	Group A	Group B	p - value*	Group A	Group B	p - value*
0	2.58 ± 1.27	2.97 ± 1.36	0.244	5.33 ± 1.32	5.90 ± 1.03	0.069
2	2.32 ± 0.61	2.48 ± 0.68	0.321	4.43 ± 0.68	4.90 ± 0.76	0.075
6	1.90 ± 0.50	2.06 ± 0.45	0.181	3.47 ± 0.57	3.77 ± 0.63	0.057
12	1.77 ± 0.50	2.03 ± 0.65	0.813	2.87 ± 0.57	3.07 ± 0.67	0.227
24	1.83 ± 0.40	1.93 ± 0.75	0.527	2.56 ± 0.63	2.83 ± 0.79	0.153

*p - value (> 0.05) non-significant at all intervals at rest as well as on movement.

the groups. This difference was not statistically significant. ($p > 0.05$) There was no incidence of urinary retention, pruritus, pneumothorax or respiratory depression in any of the group. 7 out of the 10 patient with Post-operative Nausea-vomiting (PONV) in group A required anti-emetic; while 5 out of 8 patients with PONV required rescue anti-emetic in group B.

Discussion

Pain is a critical focus of patient care. Substantial improvement in knowledge of mechanisms and treatment of pain has been outcome of extensive research, but unfortunately, this has not been translated into appropriate patient satisfaction. Post-operative pain is still inadequately relieved. This study focusing on alleviating the acute post-operative pain following MRM by performing PVB and prolonging this duration of analgesia by addition of an adjuvant in form of Ketamine along with local anesthetics in PVB.

The demographic parameters of the patients included in both the groups were comparable in this study. The duration of surgery in the both groups was also comparable. The variation in duration of surgery among these patients could be attributed to the varied skill and expertise of the operative surgeon and intra-operative finding.

In present study, paravertebral block was combined with GA. For the same reason 0.25% Bupivacaine was used instead of 0.5%. Burlacu *et al.* too had combined single shot paravertebral block (0.25% bupivacaine) with GA. Paravertebral space is not as isolated structure but communicates with paravertebral space above and below.[9] Thus, in this study USG guided technique was performed instead of blind technique, this was to ensure an increased probability of successful block. The block was performed just prior to induction of anesthesia. The onset of block quoted in various studies varies from *10–20 minutes*.[10] The sensory block however, could not be checked as general anesthesia was induced immediately following block.

Single injection PVB at T_4 level was found to be a suitable alternative to GA in women undergoing breast surgery by Pusch *et al.*[11] The multisegmental spread of single injection paravertebral block was confirmed by Saito *et al.* in a voluntary study.[12] Burlacu *et al.* also confirmed the efficacy of single injection paravertebral block at T_4 level.[6]

One of ways in which the efficacy of block can be assessed is checking for sensory loss for pin prick, which could not be assessed in this study as GA was immediately induced after performing block. But as a surrogate to checking of sensory loss, intra-operative analgesic requirement was used. The mean fentanyl consumption in groups A and B were comparable. The difference was statistically non-significant. About 60 % in group A and 63.33 % in group B patients did not require fentanyl intra-operatively. Thus, block was fully effective in these patients. In the rest of the patients the block was either partially effective or failed.

Moore *et al.* described that there is a tendency for caudal spread of the drugs when injected into paravertebral space.[13] This explain inadequate block at T_1 dermatome in this study, as the block was performed at T_4 level. In this study, sitting posture was used for performing block, which could not influenced spread of drugs in TPVS.

The VAS scores at rest in the immediate post-operative period were comparable in the both groups. This might be because of analgesia provided by block and fentanyl supplementation provided intra-operatively, which continued with PCA morphine in the immediate post-operative periods in the both groups. This is similar to observation made in other studies which showed low pain scores in immediate post-operative period.[6] The subsequent VAS scores on movement were also comparable in both the groups. This was because of the participants had already been instructed to call nurse to inject morphine in order to maintain their

pain score less than 4. VAS score greater than 3 was considered the cut off for inadequate analgesia based on several studies reviewed by Dolin *et al*.[14]

The efficacy of block was also assessed with morphine consumption in post-operative period in the both groups. First requirement of rescue analgesia in post-operative period were similar in both the groups and difference was statistically insignificant. Similar result was found by Singh *et al*.[15] In this study, mean total consumption of morphine in group A and group B were also similar. This difference was statistically non-significant. This could be attributed to the efficacy of block being similar in the both groups.

In this study, we did not find any prolongation in duration of block. This variable effect of ketamine probably can be explained from different site of injection. In human study, showing effective analgesia, ketamine with local anesthetics was administered with incisional infiltration of subcutaneously.[10,16] The analgesic effect thus may have been consequences of a pure local effects of ketamine at the level of surgical trauma where a wound inflammation occurs.[17,18]

The dose of ketamine used (*0.5 mg/kg*), might have been absorbed quickly in systemic circulation and any local anesthetic effects could have been masked especially with the long acting used Bupivacaine local anesthetic. This also might have happened in Lee *et al.* study as they injected their study solution in the interscalene area which is vessels rich.[19] The relative high incidence of ketamine related psychomimetic adverse effects in this study may support this explanation. In present study, no psychomimetic effect were seen any of the groups. This can be explained as the good analgesia enhancing effect and lack of psychomimetic effect of ketamine when given in the epidural or caudal route where the systemic absorption is slow. The present study hypothesized that ketamine either act at the nerves as they emerges from intervertebral foramen or diffuse into epidural space and act on spinal cord. However, results obtained in this study do not substantiate either of these hypotheses.

In this study, there was no episode of intra-operative hypotension in either of two groups. This could be due to intravascular absorption of ketamine from paravertebral space. The absorption from paravertebral space is quite high, ranked next only to that from the intercostal nerve block.[20] Ketamine stimulates cardiovascular system and usually associated with increases in HR and BP.[21] The post-operative hemodynamic parameters were also comparable between both the groups. This could be because of maintaining lower pain scores in both the groups.

The PVB technique is associated with certain complication like pleural puncture, pneumothorax, epidural or subarachnoid placement, intravascular injection and horner's syndrome.[22] No incidence of pneumothorax was observed in this study. This might be attributed to the enhanced safety associated with USG Guided technique and less number of cases. Several others studies on PVB for breast surgery show similar results.[23,24]

Epidural or subarachnoid spread has also been reported with PVB. Weltz *et al.* have reported 2 cases of epidural spread of local anesthetics in a study of thirty patients using PVB as the sole anesthesia for inguinal hernia repair.[25] Klein *et al.* also reported one incidence of epidural spread without hemodynamic in stability out of 24 patients receiving PVB for inguinal herniorrhaphy.[26] The present study also, did not have any case of epidural spread.

Incidence of failed block could not be estimated from this study as we did not assess loss of sensation following PVB placement and general anesthesia was induced in most patients immediately after PVB. Failure rate after PVB in adults varies from 6.1–10.7%.[23] This reflects technically difficulty in identifying paravertebral space. The above quoted figures are failure rate following nerve stimulator guided technique. Several others studies on PVB for breast surgery quote similar failure rates.

The mean PONV scores immediately post-operative period were comparable in the both groups. In this study, similar PONV scores and incidence between the two groups could be because of combining PVB with GA. This observation has again confirmed by other studies.[11,25,27]

Conclusion

The present study showed that the addition of ketamine to bupivacaine did not improve the efficacy or duration of paravertebral analgesia in the post operative and intra-operative period in patients undergoing modified radical mastectomy.

References

1. Merskey H, Bugduk N. Classification of chronic pain. Description of chronic pain syndrome and definitions of pain terms, 2nd edition. Seattle, WA: IASP Press; 1994. pp. 180–96.

2. Perkins F, Kehlet H. Chronic pain as an outcome of surgery: A review of predictor factors. Anesthesiology. 2000;93(4):1123–33.
3. Roy Greengrass, R Weltz Christina, Dirk Iglehart J. Use of paravertebral block anesthesia in Surgical Management of Breast Cancer. Annals of Surgery. 1998;227:496–501.
4. Eason MJ, Waytt R. Paravertebral thoracic block: A Reappraisal Anesthesia. 1979;34:638–642.
5. Gilbert J, Huntman J. Thoracic paravertebral block: A method of pain con. Acta Anesthesiol Scand. 1989;33:142–45.
6. Burlacu CL, Frizelle HP, Moriarty DC. Fentanyl and clonidine as adjuvant analgesics with levobupivacaine in paravertebral analgesia for breast surgery. Anesthesia. 2006;61:932–37.
7. Abdel Ghaffar ME, Abdulatif M, Al-Gandhi A, *et al*. Epidural ketamine reduces post-operative epidural PCA consumption of fentanyl/ bupivacaine. Can J Anesth. 1998;45:103–109.
8. Himmelseher S, Ziegler-Pithamitsis D, Argiriadou H, *et al*. Small dose S-ketamine reduces post-operative pain when applied with ropivacaine in epidural anesthesia for total knee arthroplasty. Anesth Analg. 2001;92:1290–295.
9. Klein SM, Nielsen KC, Ahmed N, *et al.* In situ images of the thoracic paravertebral space. Reg Anesth Pain Med. 2004;29:596–99.
10. Martindale SJ, Dix P, Stoddart PA. Double blind randomized controlled trial of caudal versus intravenous S (+) ketamine for supplementation of caudal analgesia in children. Br J Anesth. 2004;92:344–47.
11. Pusch F, Freitag H, Weinstabl C. Single injection paravertebral block compared to general anesthesia for breast surgery. Acta Anesthesiol Scand. 1999;43:770–74.
12. Saito T, Den S, Cheema SPS. A single injection multisegmental paravertebral block extension of somatosensory and sympathetic block in volunteers. Acta Anesthesiol Scand. 2001;45: 30–33.
13. Moore DC. Intercostals nerve block: Spread of Indian ink injected into the subcostal groove. Br J Anesth. 1981;53:325.
14. Dolin SJ, Cashman JN, Bland JM. Effectiveness of acute post-operative pain management I. Evidence from published data. Br J Anesth. 2002;89:409–23.
15. Singh A, Kushwawa JK, Gupta R, *et al*. A comparative study between morphine, dexmedetomidine and ketamine as an adjunct to levobupivacaine in paravertebral block during modified radical mastectomy. Indian J of Research. 2016;10:27–31.
16. De Negri P, Ivani G, Visconti C, *et al*. How to prolong post-operative analgesia after caudal anesthesia with ropivacaine in children: S-ketamine versus clonidine. Pediatr Anesth. 2001;11:679–83.
17. Weber WV, Jawalekar KS, Jawalekar SR. The effect of ketamine on the nerve conduction in isolated sciatic nerve of toad. Neurosci Lett. 1975;1:115–20.
18. Tverskoy M, Oren M, Vaskovich M, *et al.* Ketamine enhances local anesthetic and analgesic effects of bupivacaine by peripheral mechanism: A study in post-operative patients. Neurosci Lett. 1996;215:5–8.
19. Lee IO, Kim WK, Kong MH, *et al*. No enhancement of sensory and motor by ketamine added to ropivacaine interscalene brachila plexus blockade. Acta Anesthesiol Scand. 2002;46:821–26.
20. Morgan Jr GE, Mikhail MS, Murray MJ. Local anesthetic. Clinical anesthesilogy, 4th edition. New York: McGraw-Hill; 2008. pp. 263–76.
21. Reves JG, Glass PSA, Lubarsky DA, *et al.* Intravenous non-opioids anesthetics. Miller's Anesthesia, 6th edition. Philadelphia: Churchill Livingstone; 2005. pp. 317–78.
22. Karmakar MK. Thoracic paravertebral block. Anesthesiology. 2001;95:771–80.
23. Lonnqvist PA, MacKenzie J, Soni AK, *et al.* Paravertebral blockade: Failure rate and complication. Anesthesia. 1995;50:813–15.
24. Klein SM, Teele SM, Greengrass RA. A clinical overview of paravertebral blockade. The Internet Journal of Anesthesiology. 1999;3:1–6.
25. Weltz CR, Greengrass RA, Lyerly HK. Ambulatory surgical management of breast carcinoma using paravertebral block. Ann Surg. 1995;222:19–26.
26. Klein SM, Pietroban R, Nielsen KC, *et al.* Paravertebral somatic nerve block compared with peripheral nerve block for outpatients inguinal herniorrhaphy. Reg Anesth Pain Med. 2002;27:476–80.
27. Moller JF, Nikolajsen L, Rodt SA, *et al.* Thoracic paravertebral block for breast cancer surgery: A randomized double blind study. Anesth Analg. 2007;105:1848–851.