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Efficacy of Levobupivacaine and Ropivacaine in Postoperative Pain control after Surgical Removal of Impacted Mandibular Third Molar: A Split Mouth Randomized Clinical Trial

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Objectives: The aim of the present clinical trial study is to compare the efficacy of 0.75% levobupivacaine with that of 0.75% ropivacaine for pain control after surgical removal of impacted mandibular third molars.
Methods: This prospective study included 40 patients (30 females and 10 males) who had been referred to the Department of Oral and Maxillofacial Surgery for surgical removal of third molars of similar difficulty index in two separate sessions under local anaesthesia. Within each patient, levobupivacaine was used to anesthetize one extraction side and for the other side, ropivacaine was used. Onset of anaesthesia, duration of surgery, timing of pain appearance and analgesic consumption were evaluated. Data collected was statistically analysed and results obtained.
Results: In this study we observed, there were no significant differences in onset of anaesthesia and duration of surgical procedure between the two groups (P> 0.05). Timing of pain appearance and of first drug consumption was earlier in the ropivacaine group than levobupivacaine group and the results were statistically significant (P< 0.05). Patients with levobupivacaine anaesthesia had significantly lower visual analogue (VAS) pain scores at 1 and 2 hours postoperatively than those with ropivacaine anaesthesia.

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Conclusion: Within the limitations of the present study, Levobupivacaine is a valid alternative to traditional local anaesthetics for surgical removal of lower third molars. It presents better pain relief when compared to ropivacaine in the immediate postoperative period as evidenced by lower VAS scores.

Keywords: Third molar; anaesthesia; levobupivacaine; ropivacaine; mandibular molar; postsurgical pain.

1. INTRODUCTION

The continuous improvement in local anaesthetic agents has contributed more than any other factor to the control of pain during and especially after dental surgery. The surgical removal of the lower third molars are a common oral surgical procedure which causes severe postoperative pain [1]. Lidocaine is the one most frequently used in dentistry, and is the benchmark for any comparison [2]. The idea that using long-lasting local anaesthetic, such as bupivacaine or its safer derivatives ropivacaine and levobupivacaine, would improve the quality of care after removal of mandibular third molars is based on the fact that a part of postoperative time would be covered by residual effects of anaesthesia, thus reducing pain or analgesics consumption [3].

Bupivacaine, the long-acting amide local anaesthetic, is used to obtain both effective sensory block with long-lasting duration and beneficial postoperative analgesia for surgical extraction of lower third molars. However, after the clinical reports of life threatening neural and cardiac toxicity of bupivacaine [4–6], it became evident that bupivacaine has a narrow safety margin given its high lipid solubility as opposed to other amide local anaesthetics [7,8]. Commercial preparations of bupivacaine exist as a racemic solution, containing equal amounts of the two enantiomers, R(+) dextrorotatory and S(−) levorotatory stereoisomers [9]. Consequently, vasoactivity and toxicity of the S(−) levorotatory and R(+) dextrorotatory enantiomers of bupivacaine differ, while levorotatory enantiomer are more vasoconstrictive and less toxic [10–12].

Ropivacaine is a new long-acting enantiomerically pure (S-enantiomer) amide local anaesthetic, structurally related to bupivacaine, but with less cardiac toxicity and neurotoxicity [13]. It is shown to be very suitable for regional anaesthesia and has been tested in dentistry with encouraging results about its duration of action [14]. Its low liposolubility blocks nerve fibers involved in pain transmission (thin Aδ and C fibers) to a greater degree than those controlling motor function (large Aβ fibers). Unlike most local anaesthetics, which are vasodilators, ropivacaine produces vasoconstriction in vitro and in vivo in animal models [15,16].The vasoconstrictive properties and strong bond to plasma proteins prolong anaesthesia duration.

Levobupivacaine, as the pure S(−) isomer, has been developed as an alternative to bupivacaine with the desirable blocking properties of racemic bupivacaine, due to a greater margin of safety. Moreover, the comparative clinical studies evaluating levobupivacaine (LBUP) for peripheral nerve blocks have suggested that clinical parameters of regional anaesthesia produced with 0.5 % levobupivacaine might be similar or even better than those produced with an equivalent dose of bupivacaine [17,18]. Levobupivacaine has been also reported to possess advantages in terms of cardiotoxicity and CNS toxicity in animal and human volunteer studies [19,20].

Previously our team has a rich experience in working on various research projects across multiple disciplines [21–35]. Now the growing trend in this area motivated us to pursue this project. Based on this inspiration, we aim to compare the efficacy of 0.75% levobupivacaine with that of 0.75% ropivacaine for pain control after surgical removal of impacted mandibular third molars.

2. MATERIALS AND METHODS

2.1 Study Setup

This randomized prospective controlled clinical study was conducted among patients reporting to the outpatient dental department of Oral Surgery clinic. The study population included 40 adult patients who were randomly selected and had been referred to the department of oral and maxillofacial surgery for surgical removal of impacted mandibular molar. A split-mouth design

was chosen. Random sampling by means of opaque, sealed envelopes was used to determine which of the two mandibular molars would be extracted first. Local anaesthetics used were levobupivacaine and ropivacaine [Fig. 1]. All extractions were performed under local anaesthesia, without any premedication, by the same surgeon using a standard technique. The second extraction was carried out 1 month later.

Group (1): Levobupivacaine group- inferior alveolar nerve block was performed by means of 2ml solution (0.75% levobupivacaine) without epinephrine. In addition, the buccal soft tissues were infiltrated with 1ml solution [Fig. 2].

Group (2): Ropivacaine group- inferior alveolar nerve block was performed by means of 2ml solution (0.75% ropivacaine) without epinephrine. In addition, the buccal soft tissues were anesthetized with 1 ml solution with 1:80.000 epinephrine [Fig. 3].

2.2 Inclusion Criteria

- Patients between 18 years-50 years of age
- Both genders
- An inflammatory state around third molar
- Patients with clinical and radiographic records requiring surgical removal of bilateral impacted mandibular 3rd molars.

2.3 Exclusion Criteria

- Patients with incomplete clinical and radiological records.
- Patients with severe systemic conditions like diabetes and hypertension.
- Patients with hypersensitivity to honey, alcoholism, drug abuse.

2.4 Procedure

A split-mouth design was chosen. Random sampling by means of opaque, sealed envelopes was used to determine which of the two mandibular molars would be surgically removed first. All extractions were performed under local anaesthesia, without any premedication, by the same surgeon using a standard technique. A mucoperiosteal flap was reflected to gain access to the impacted third molar. Thereafter, bone was removed by a water-cooled bur in a surgical drill. All wounds were closed with 3-0 polyglactin (Vicryl) interrupted sutures. The duration of each operation from incision to wound closure was recorded. Both local anaesthetics, levobupivacaine and ropivacaine, were tested on the same patient. For the first extraction, the choice of the anaesthetic was randomized and performed in blocks of four patients by means of an opaque, sealed envelope. Therefore, each patient served as his or her own control. The second extraction was performed 1 month later using the anaesthetic not used for the first extraction. Therefore, each patient was blind to the anaesthetics type.

2.5 Study Parameters

The following data were extracted for the purpose of the study:

- Age of the patient
- Gender of the patient
- Intraoperative period- Onset of anaesthesia: defined as the period between the end of the local anaesthetic administration and the onset of lower lip anaesthesia;
- Intraoperative period- Duration of surgery; from incision to wound closure
- Post operative VAS pain Scores at 1st, 2nd, 12th and 24th hour.
- Time lapse to postoperative pain;
- Time lapse to first analgesic intake

All patients were asked by a single person blind to the anaesthetic used to complete a diary reporting on the last three parameters for 24 hours after surgery. These data were recorded after administration of both levobupivacaine and ropivacaine.

2.6 VAS Score

Patients were asked to record the pain intensity on a visual analogue scale (VAS) with the anchor points “0 = no pain” and “10 = the worst pain imaginable.” The VAS is a sensitive and reliable method for recording pain intensity and is considered to be better than the verbal, digital, numerical, and descriptive scales [36]. The subjects were divided into four age groups- Group 1: 11-20 years, Group 2: 21-30 years, Group 3: 31-40 years, Group 4: 41-50 years.

2.7 Data Collection

The data relating to the study parameters were obtained from among patients who reported to the Outpatient Department of Oral and Maxillofacial Surgery. Approval for the study was obtained from the Institutional Ethical Committee. All assessments were done by a single examiner

and the findings were reviewed and recorded by two investigators. Each patient gave his or her written, informed consent to participate and had the right to withdraw from the trial at any time.

2.8 Statistical Analysis

The data were tabulated and analysed using IBM SPSS version 23.0 software. Descriptive statistics were expressed by mean, standard deviation and frequency, percentage based on the type of data. Student's t-test was used to compare variables (Onset of Anaesthesia and Duration of Surgery, Timelapse to postoperative pain and 1st rescue analgesic intake) between Levobupivacaine and Ropivacaine Groups. The effects over time of the two anaesthetics on pain intensity were evaluated by analysis of variance (ANOVA). The significance level was set at $P<0.05$ with a confidence interval of 95%.

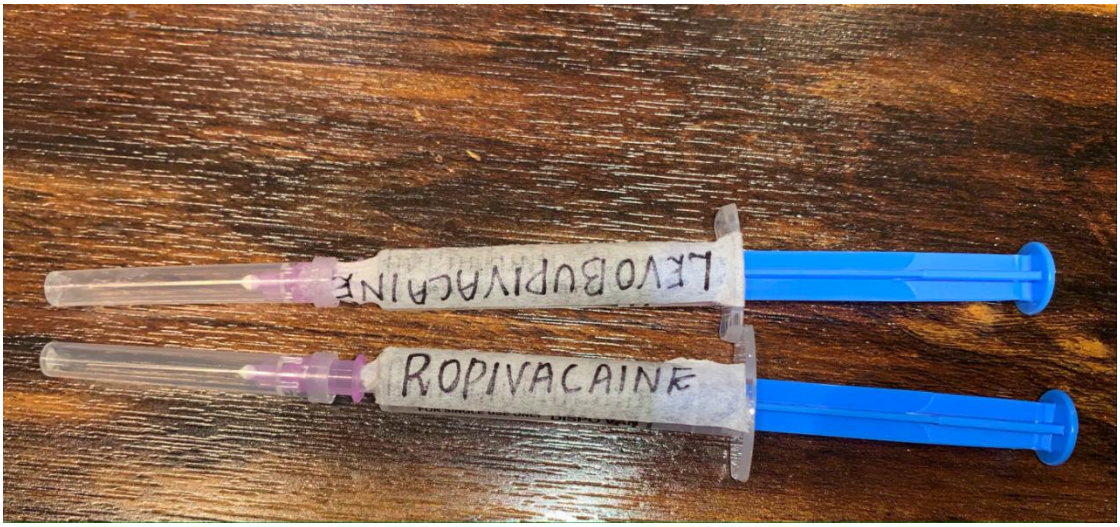

Fig. 1. Levobupivacaine and Ropivacaine Injection

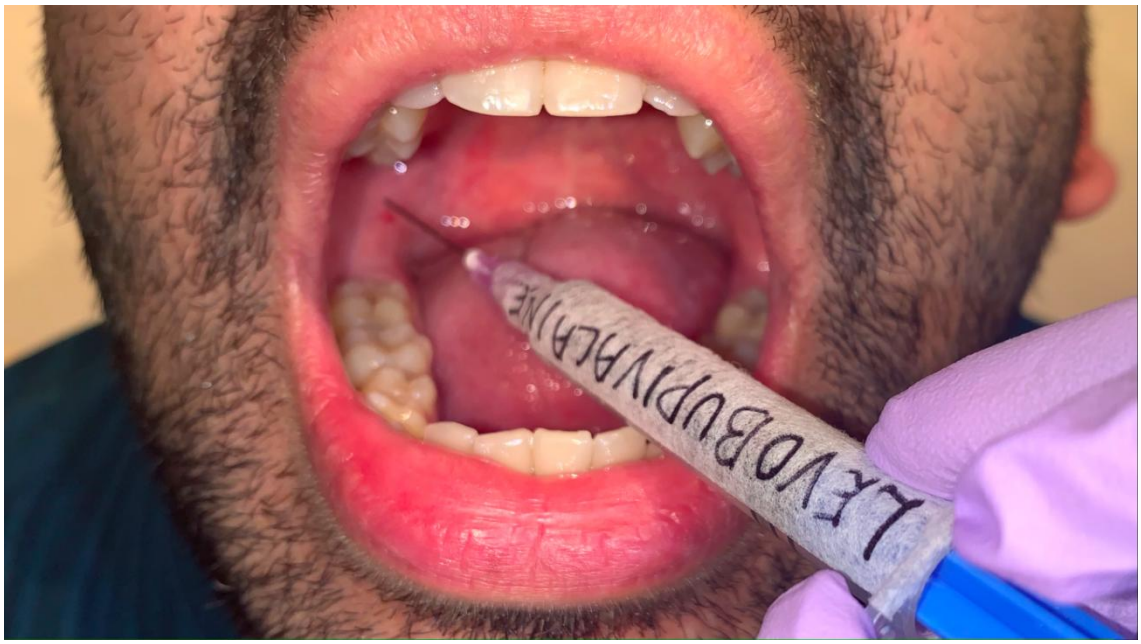

Fig. 2. Nerve block performed with Levobupivacaine.

In the Levobupivacaine group, an inferior alveolar nerve block was performed by means of 2ml solution (0.75% levobupivacaine) without epinephrine. In addition, the buccal soft tissues were infiltrated with 1.0 ml

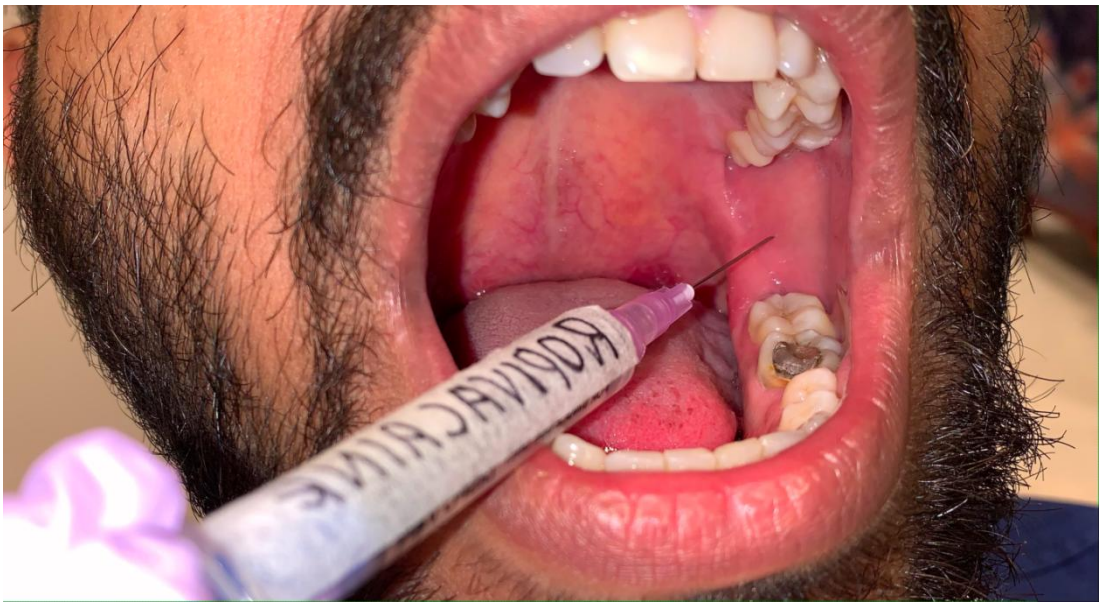

Fig. 3. Nerve block performed with Ropivacaine

In the Ropivacaine Group, an Inferior alveolar nerve block was performed by means of a 2ml solution (0.75% ropivacaine) without epinephrine. In addition, the buccal soft tissues were infiltrated with 1.0 ml

3. RESULTS

A total of 40 patients participated in this study, with an overall 100% participation.

3.1 Age Distribution

The youngest and oldest patients were aged 18 and 50 years, respectively. The distribution of study subjects based on age revealed that most patients belonged to the 31-40 years of age group. (67.50%)

3.2 Gender Distribution

The distribution of study subjects based on gender, over a ten-month period, revealed that 30 patients (75%) women and 10 patients (25%) men participated in this study.

3.3 Onset of Anaesthesia and Duration of Surgery

There was no statistically significant difference in onset time of anaesthesia (P=0.07) and duration of surgery (P=0.278) between the two groups.

3.4 Post Operative Pain Evaluation by Visual Analogue Scale

Three patients after levobupivacaine anaesthesia and two patients after ropivacaine anaesthesia did not feel pain during the 24 hours post-surgery. Among all patients with postoperative pain, the timing of pain appearance was significantly longer after levobupivacaine than ropivacaine anaesthesia. Pain score at 1st and 2nd hours after surgery were different between the two anaesthetics; the mean VAS scores recorded after injection of levobupivacaine at 1 and 2 hours respectively were significantly lower than after ropivacaine at 1 and 2 hours, respectively (Fig. 4). No significant differences in pain scores were observed between the two anaesthetics at 12th and 24 hours post-surgery (P>0.05).

3.5 Time Lapse to Postoperative Pain and 1st Rescue Analgesic Intake

Eight patients after levobupivacaine and one patient after ropivacaine did not need rescue analgesia for postoperative pain. In patients who required analgesia, the timing of the first drug intake was significantly longer for levobupivacaine than ropivacaine (P<0.05) [Table 1].

There was no statistically significant difference in the onset time of anaesthesia (P=0.07) and duration of surgery (P=0.278) between the two groups. In patients who required analgesia, the timing of pain appearance and the first analgesic drug intake was significantly longer for levobupivacaine than ropivacaine and the results were statistically significant (P<0.05).

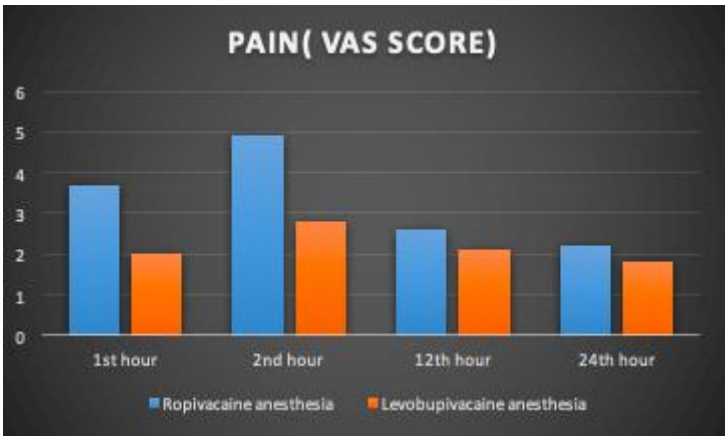

Fig. 4. Bar diagram depicts VAS scores of the ropivacaine group (blue) and the levobupivacaine group (orange) at the 1st, 2nd, 12th and 24th-hour post-surgery

The X-Axis depicts the Post extraction hour and Y-Axis represents the VAS Scores. The VAS scores of the ropivacaine group were higher than the levobupivacaine group

Table 1. Demonstrating the distribution of variables between the two study groups

Variable	Ropivacaine Group (Mean)	Levobupivacaine Group (Mean)	Test Value	P Value
Onset of Anaesthesia (mins)	82	114	-9.00	0.07
Surgery Time (mins)	25	29	-2.143	0.278
Time lapse to postoperative pain (mins)	180	320.5	-36.3	0.018*
Time lapse to first analgesic intake (mins)	202	420	-23.4	0.027*

*Students t test; * statistically significant at p<0.05*

4. DISCUSSION

Long-acting local anaesthetic agents are suggested in lengthy dental treatments for the control of post-surgical pain. Although the available anaesthetic for dentistry has minimal side effects in the doses usually employed, they might cause potential problems. Bupivacaine, for example, shows toxic effects on the central nervous and cardiovascular systems, while etidocaine may affect intraoperative bleeding [3,36].

The present study demonstrated that levobupivacaine is more effective than ropivacaine for pain control in the first hours after dental extraction. Also, the delay to pain appearance and to the first analgesic needed was longer in patients receiving levobupivacaine than ropivacaine. The absence of significant differences in pain intensity in the following VAS evaluations (12th and 24 hours) may be related to the use of systemic analgesic, although it should be noted that fewer patients needed an analgesic after levobupivacaine than ropivacaine anaesthesia.

The prolonged duration of action may be attributable to the intrinsic characteristics of levobupivacaine presenting higher lipid solubility and protein-binding properties than ropivacaine. This result is in accordance with that of a previous study by Crincoli et al [37], in which the use of levobupivacaine was associated with both significantly longer analgesia duration and time-lapse to rescue analgesia when compared to ropivacaine. In another study performed by Brajkovic et al [9], levobupivacaine was shown to produce a more effective surgical block when the intensity of the mandibular nerve block was measured by VAS Scale. Also, the time of first postoperative pain report and time of first

analgesic dose taken was significantly prolonged in the levobupivacaine group. This is also in accordance with the results of our study.

Rood and co-workers [38] demonstrated the use of levobupivacaine in oral surgery where they compared the efficacy of 0.75% levobupivacaine (without vasoconstrictor) with 2% lignocaine (with adrenaline 1:80,000) and with placebo for postoperative pain control in 93 patients who underwent removal of mandibular third molars under general anaesthesia. Their results are similar to our study regarding the lower number of patients requiring rescue analgesia, the lower pain scores, and the larger time laps to analgesic intake after levobupivacaine than ropivacaine.

However, there are differences in the design of the two studies. In our study, levobupivacaine versus ropivacaine was tested and surgery was performed only by one surgeon while Rood and co-workers [38] compared levobupivacaine versus lignocaine and placebo and surgery was performed by two surgeons. Also, in the study performed by Rood and co-workers [38], subjects underwent oral surgery under general anaesthesia while in our study, subjects were operated under local anaesthesia with no premedication. Patients under local anaesthesia, being awake and conscious, could provide details on the onset of the anaesthesia and what they felt immediately after surgery.

Also, patients in our study did not get pre-medications while in the earlier study two patients in the levobupivacaine, three in the lignocaine, and seven in the placebo groups received fentanyl, a powerful opioid analgesic that could have further influenced the results. All patients in the study were discharged the same day of surgery after receiving a supply of analgesics (ibuprofen), while patients of our study took analgesics only in case of need.

Finally, in both studies, the pain was evaluated with VAS. However, in the present study, a split-mouth design was used such that each patient was his or her own control, while Rood [38] and co-workers divided subjects into three groups: placebo, lignocaine, and levobupivacaine. On assessing the onset, no difference in the onset and quality (i.e, how the patients responded to the drug) of anaesthesia between ropivacaine and levobupivacaine was observed. This was also in accordance with the study performed by Crincoli et al [1]. Our institution is passionate about high-quality evidence-based research and has excelled in various fields [25,39–58].

5. CONCLUSION

Within the limits of this study, it can be concluded that differences between two anaesthetics were seen in the postoperative pain intensity, prolonged time of first postoperative pain report and time of first analgesic consumption. Thus, levobupivacaine is a valid alternative to traditional local anaesthetics for surgical removal of lower third molars. It presents better pain relief when compared to ropivacaine in the immediate postoperative period as evidenced by lower VAS scores.

6. LIMITATIONS OF THE STUDY

As the VAS Scores were based on patients' perception, a subjective opinion regarding the results was obtained. The pain threshold for different patients would not be similar. However, Also, further studies are needed to compare the safety profile of levobupivacaine and ropivacaine during mandibular nerve blocks in oral surgery.

7. FUTURE SCOPE

Levobupivacaine presents clinical advantages when compared to ropivacaine for postoperative pain control after surgical removal of impacted mandibular third molars and may therefore be considered a valid and safe alternative to traditional local anaesthetics.

CONSENT

As per international standard or university standards, participants' written informed consent has been obtained and preserved by the author(s).

ETHICAL APPROVAL

Ethical approval was obtained from the institution's ethical committee.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Crincoli V, Favia G, LImongelli L, Tempesta A, Brienza N. The effectiveness of ropivacaine and mepivacaine in the postoperative pain after third lower molar

surgery. International Journal of Medical Sciences. 2015;12(11):862.
2. Bouloux GF, Punnia-Moorthy A. Bupivacaine versus lidocaine for third molar surgery: a double-blind, randomized, crossover study. J Oral Maxillofac Surg. 1999;57:510–4..
3. Crincoli V, Favia G, LImongelli L, et al. The Effectiveness of Ropivacaine and Mepivacaine in the Postoperative Pain after Third Lower Molar Surgery. Int J Med Sci. 2015;12:862–866.
4. Brown DL, Ransom DM, Hall JA, et al. Regional Anesthesia and Local Anesthetic-Induced Systemic Toxicity. Anesthesia & Analgesia. 1995;81:321–328.
5. Kopp SL, Wynd KP, Horlocker TT, et al. Regional blockade in patients with a history of a seizure disorder. Anesth Analg. 2009;109:272–278.
6. Marwick PC, Levin AI, Coetzee AR. Recurrence of cardiotoxicity after lipid rescue from bupivacaine-induced cardiac arrest. Anesth Analg. 2009;108:1344–1346.
7. Leone S, Di Cianni S, Casati A, et al. Pharmacology, toxicology, and clinical use of new long acting local anesthetics, ropivacaine and levobupivacaine. Acta Biomed. 2008;79:92–105.
8. Werdehausen R, Braun S, Fazeli S, et al. Lipophilicity but not stereospecificity is a major determinant of local anaesthetic-induced cytotoxicity in human T-lymphoma cells. Eur J Anaesthesiol. 2012;29:35–41.
9. Brajkovic D, Brkovic B, Milic M, et al. Levobupivacaine vs. bupivacaine for third molar surgery: quality of anaesthesia, postoperative analgesia and local vascular effects. Clin Oral Investig. 2014;18:1481–1488.
10. Aps C, Reynolds F. An intradermal study of the local anaesthetic and vascular effects of the isomers of bupivacaine. Br J Clin Pharmacol. 1978;6:63–68.
11. Vanhoutte F, Vereecke J, Verbeke N, et al. Stereoselective effects of the enantiomers of bupivacaine on the electrophysiological properties of the guinea-pig papillary muscle. British Journal of Pharmacology. 1991;103:1275–1281.
12. Mazoit JX, Kantelip J-P, Orhant EE, et al. Myocardial uptake of lignocaine: pharmacokinetics and pharmacodynamics in the isolated perfused heart of the rabbit. British Journal of Pharmacology. 1990;101: 843–846.
13. Hansen TG. Ropivacaine: A pharmacological review. Expert Rev Neurother. 2004;4:781–791.
14. Davis MJ, Vogel LD. Local anesthetic safety in pediatric patients. N Y State Dent J. 1996;62:32–35.
15. Iida H, Ohata H, Iida M, et al. The differential effects of stereoisomers of ropivacaine and bupivacaine on cerebral pial arterioles in dogs. Anesth Analg 2001; 93:1552–6.
16. Timponi CF, Oliveira NE, Arruda RMP, et al. Effects of the local anaesthetic ropivacaine on vascular reactivity in the mouse perfused mesenteric arteries. Basic Clin Pharmacol Toxicol. 2006;98:518–520.
17. Cox CR, Checketts MR, Mackenzie N, et al. Comparison of S(-)-Bupivacaine with Racemic (RS)-Bupivacaine in Supraclavicular Brachial Plexus Block. Survey of Anesthesiology. 1999;43:172–173.
18. Casati A, Chelly JE, Cerchierini E, et al. Clinical properties of levobupivacaine or racemic bupivacaine for sciatic nerve block. J Clin Anesth. 2002;14:111–114.
19. Huang YF, Pryor ME, Mather LE, et al. Cardiovascular and central nervous system effects of intravenous levobupivacaine and bupivacaine in sheep. Anesth Analg. 1998;86:797–804.
20. Bardsley H, Gristwood R, Baker H, et al. A comparison of the cardiovascular effects of levobupivacaine and rac-bupivacaine following intravenous administration to healthy volunteers. British Journal of Clinical Pharmacology. 1998;46:245–249.
21. Govindaraju L, Gurunathan D. Effectiveness of Chewable Tooth Brush in Children-A Prospective Clinical Study. J Clin Diagn Res. 2017;11:ZC31–ZC34.
22. Christabel A, Anantanarayanan P, Subash P, et al. Comparison of pterygomaxillary dysjunction with tuberosity separation in isolated Le Fort I osteotomies: a prospective, multi-centre, triple-blind, randomized controlled trial. Int J Oral Maxillofac Surg. 2016;45:180–185.
23. Soh CL, Narayanan V. Quality of life assessment in patients with dentofacial deformity undergoing orthognathic surgery--a systematic review. Int J Oral Maxillofac Surg. 2013;42:974–980.
24. Mehta M, Deeksha, Tewari D, et al. Oligonucleotide therapy: An emerging focus area for drug delivery in chronic

inflammatory respiratory diseases. Chem Biol Interact. 2019;308:206–215.
25. Ezhilarasan D, Apoorva VS, Ashok Vardhan N. Syzygium cumini extract induced reactive oxygen species-mediated apoptosis in human oral squamous carcinoma cells. J Oral Pathol Med. 2019; 48:115–121.
26. Campeau PM, Kasperaviciute D, Lu JT, et al. The genetic basis of DOORS syndrome: an exome-sequencing study. Lancet Neurol 2014;13:44–58.
27. Kumar SSS. Knowledge and awareness regarding antibiotic prophylaxis for infective endocarditis among undergraduate dental students. Asian J Pharm Clin Res. 2016;154.
28. Christabel SL. Prevalence of type of Frenal Attachment and morphology of frenum in children, Chennai, Tamil Nadu. World J Dent 2015; 6: 203–207.
29. Kumar S, Rahman R. Knowledge, awareness, and practices regarding biomedical waste management among undergraduate dental students. Asian J Pharm Clin Res 2017;10:341.
30. Sridharan G, Ramani P, Patankar S. Serum metabolomics in oral leukoplakia and oral squamous cell carcinoma. J Cancer Res Ther. 2017;13:556–561.
31. Ramesh A, Varghese SS, Doraiswamy JN, et al. Herbs as an antioxidant arsenal for periodontal diseases. J Intercult Ethnopharmacol. 2016;5:92–96.
32. Thamaraiselvan M, Elavarasu S, Thangakumaran S, et al. Comparative clinical evaluation of coronally advanced flap with or without platelet rich fibrin membrane in the treatment of isolated gingival recession. J Indian Soc Periodontol 2015;19:66–71.
33. Thangaraj SV, Shyamsundar V, Krishnamurthy A, et al. Molecular Portrait of Oral Tongue Squamous Cell Carcinoma Shown by Integrative Meta-Analysis of Expression Profiles with Validations. PLoS One 2016;11:e0156582.
34. Ponnulakshmi R, Shyamaladevi B, Vijayalakshmi P, et al. In silico and in vivo analysis to identify the antidiabetic activity of beta sitosterol in adipose tissue of high fat diet and sucrose induced type-2 diabetic experimental rats. Toxicol Mech Methods 2019;29:276–290.
35. Ramakrishnan M, Shukri M. Fluoride, Fluoridated Toothpaste Efficacy And Its Safety In Children-Review. International Journal of Pharmaceutical Research. 2018;10(04):109-14.
36. Tuffin JR, Cunliffe DR, Shaw SR. Do local analgesics injected at the time of third molar removal under general anaesthesia reduce significantly post operative analgesic requirements? A double-blind controlled trial. British Journal of Oral and Maxillofacial Surgery. 1989;27(1):27-32.
37. Crincoli V, Bisceglie MB, Massaro M, Giuliani R, Favia G, Brienza N. Postoperative pain relief after surgical removal of impacted third molars: a single-blind, randomized, controlled study to compare levobupivacaine and mepivacaine. Journal of orofacial pain. 2009;23(4).
38. Rood JP, Coulthard P, Snowdon AT, et al. Safety and efficacy of levobupivacaine for postoperative pain relief after the surgical removal of impacted third molars: a comparison with lignocaine and adrenaline. Br J Oral Maxillofac Surg. 2002;40:491–496.
39. Vijayashree Priyadharsini J. In silico validation of the non-antibiotic drugs acetaminophen and ibuprofen as antibacterial agents against red complex pathogens. J Periodontol 2019;90:1441–1448.
40. Pc J, Marimuthu T, Devadoss P. Prevalence and measurement of anterior loop of the mandibular canal using CBCT: A cross sectional study. Clin Implant Dent Relat Res. https://europepmc.org/article/med/29624863 (2018).
41. Ramesh A, Varghese S, Jayakumar ND, et al. Comparative estimation of sulfiredoxin levels between chronic periodontitis and healthy patients - A case-control study. J Periodontol 2018;89:1241–1248.
42. Ramadurai N, Gurunathan D, Samuel AV, et al. Effectiveness of 2% Articaine as an anesthetic agent in children: randomized controlled trial. Clin Oral Investig 2019;23: 3543–3550.
43. Sridharan G, Ramani P, Patankar S, et al. Evaluation of salivary metabolomics in oral leukoplakia and oral squamous cell carcinoma. J Oral Pathol Med 2019;48: 299–306.
44. Mathew MG, Samuel SR, Soni AJ, et al. Evaluation of adhesion of Streptococcus mutans, plaque accumulation on zirconia and stainless steel crowns, and surrounding gingival inflammation in

primary molars: Randomized controlled trial. Clin Oral Investig 2020;1–6.
45. Samuel SR. Can 5-year-olds sensibly self-report the impact of developmental enamel defects on their quality of life? Int J Paediatr Dent 2021;31:285–286.
46. R H, Hannah R, Ramani P, et al. CYP2 C9 polymorphism among patients with oral squamous cell carcinoma and its role in altering the metabolism of benzo[a]pyrene. Oral Surgery, Oral Medicine, Oral Pathology and Oral Radiology 2020;130: 306–312.
47. Chandrasekar R, Chandrasekhar S, Sundari KKS, et al. Development and validation of a formula for objective assessment of cervical vertebral bone age. Prog Orthod 2020;21:38.
48. Vijayashree Priyadharsini J, Smiline Girija AS, Paramasivam A. In silico analysis of virulence genes in an emerging dental pathogen A. baumannii and related species. Arch Oral Biol 2018;94:93–98.
49. MP SK. Knowledge, Attitude and practices regarding needlestick injuries among dental students. Asian J Pharm Clin Res. 2016;9(4):312-5.
50. SK M. Knowledge, attitude, and practices regarding infection control among undergraduate dental students. Asian J Pharm Clin Res. 2016;9(1):220-4.
51. Ak H. Knowledge and awareness about oral cancer among undergraduate dental students. Asian Journal of Pharmaceutical and Clinical Research. 2016;165-7.
52. Gayathri MM. Knowledge and awareness among patients about dental implants. Journal of Pharmaceutical Sciences and Research. 2016 May 1;8(5):351.
53. Vijayalakshmi B, Kumar MS. Knowledge of students about Local anaesthetics used during oral surgical procedures. Journal of Pharmaceutical Sciences and Research. 2015;7(11):1011.
54. Gayathri MM. Knowledge, Awareness and Attitude among dental students about hepatitis B infection. Journal of Pharmaceutical Sciences and Research. 2016;8(3):168.
55. Ahamed A, Kumar MS. Knowledge, attitude and perceived confidence in handling medical emergencies among dental students. Journal of Pharmaceutical Sciences and Research. 2016;8(7):645.
56. Kumar S. Knowledge, attitude and practices of dental students toward dental management of patients on antiplatelet therapy. Asian J Pharm Clin Res. 2016; 9(30):270-6.
57. MP SK. Local hemostatic agents in the management of bleeding in oral surgery. Asian J Pharm Clin Res. 2016;9(3):35-41.
58. Kumar MP. Newer methods of extraction of teeth. Int J Pharm Bio Sci. 2015;6(3):679-85.

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