Systemic Enzymes and their Role in the Reduction of Post-Operative Edema After Surgical Removal of Lower Third Molars: A Randomized Controlled Double-Blind Study

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Abstract
Objectives: The most common postoperative complications after surgical removal of the impacted mandibular third molar tooth are edema, pain, and trismus. Different types of analgesics and anti-inflammatory agents have been employed to the reduction of these complications. The study aimed to evaluate the effect of systemic enzymes in controlling post-operative edema, pain, and trismus after surgical removal of impacted lower third molars. Methods: The enzyme combination of trypsin, bromelain, and rutoside trihydrate was compared with that of serratiopeptidase to control post-operative sequelae following lower third molar surgery. Sixty-eight patients, who required surgical removal of impacted mandibular third molars participated in this study. They were randomly allocated into two groups (n=34 in each group). Group I received 5mg serratiopeptidase orally along with the standard medication and the other group received the enzyme combination with the standard medication. P-value <0.05 was considered to be statistically significant. Results: Five patients were lost to follow-up. The mean swelling measures were found to be non-significant when compared between the two groups (Group I vs. II-54.7 ± 3.4 Vs. 53.2 ± 3.2, p=0.34). Similarly, the differences in mean pain and mouth opening values between the study groups were found to be non-significant (p>0.05). Conclusion: There was no significant difference in the effect of the enzyme combination of rutoside, trypsin, and bromelain when compared to serratiopeptidase in the reduction of postoperative edema, pain, and trismus following lower third molar surgery.
Keywords: Systemic enzymes, third molar surgery, post-operative complications, pain, edema

1. Introduction

The presence of impacted teeth in the mandible can cause problems to the adjacent teeth and mucosa if they do not erupt into the proper position. Srivastava N et al in their study concluded that impacted mandibular third molars cause distal caries in the second molars in 55% of the cases [1]. It can also lead to pericoronitis which may proceed to become a space infection involving the pterygomandibular space and considering all of the above issues with an impacted third molar, it is generally recommended to be extracted.

Halpern et al. grouped the postoperative morbidities into complications and immediate postoperative tissue reactions [2]. The latter includes swelling, pain, reduced masticatory capability, dysphagia, and trismus. The complications are unpredicted reactions that may or may not occur in the immediate postoperative time which include bleeding, dry

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socket, delayed healing, periodontal pocketing, nerve injury, and infection. The main goal should be to prevent/reduce these post-operative issues, and a variety of medications and modalities are employed including soft laser application and cryotherapy [3,4]. In the existing literature, the medical management of post-operative edema is confined mainly to the use of non-steroidal anti-inflammatory drugs (NSAIDs) or steroids. The use of enzyme treatment including trypsin, bromelain, and rutoside trihydrate for edema control in third molar surgery is not well established. Antioxidant, anti-inflammatory, analgesic, antiplatelet, and immunological modifying effects are all present in this combination [5]. In several clinical studies combination of trypsin, bromelain and rutoside has shown to have significant anti-inflammatory effects [6]. Our team has extensive knowledge and research experience that has translated into high quality publications (Pandian et al. 2018; Vikram et al. 2017; Wu et al. 2019; Sharma et al. 2019; Venu et al. 2019; Ramamurthy and Mg 2018; Samuel et al. 2020; Choudhari and Thenmozhi 2016; Ravi et al. 2017; Hannah et al. 2018; Gupta et al. 2018; Govindaraju et al. 2017; Ramesh et al. 2019; Kavarthapu and Thamaraiselvan 2018; Ashok and Ganapathy 2019)

The present study was designed to evaluate the efficacy of systemic enzymes to control postoperative edema after surgical removal of impacted mandibular third molars to explore more about this combination.

2. Materials and Methods

This study was performed in the Oral and Maxillofacial Surgery Department of Saveetha Dental College and Hospitals, Chennai. Sixty-eight patients who required surgical removal of impacted mandibular third molars were included in the study. They were randomly divided into two groups (Group I- Serratiopeptidase, Group II- Enzyme combination) of thirty-four subjects each using a random allocation software [7]. This study was reviewed and approved by the Institutional Review Board and informed consent was obtained from all the participants. The inclusion criteria were patients who were healthy individuals, aged 18 years and above, only third molars which were graded as Grade II and above in the Parant's difficulty scale, patients who had bilateral impacted teeth for whom only one side was removed surgically were included [8]. The patients with a positive medical history, those who had consumed analgesics within 24 hours before surgery, and pregnant or lactating women were excluded from this study.

Group I received 5mg serratiopeptidase orally along with the standard medication (500mg amoxicillin and 500mg paracetamol thrice daily) whereas Group II received the systemic enzyme combination with the standard medication. The proteolytic enzyme combinations used were rutoside, trypsin, and bromelain. These three agents were given in the form of an enteric-coated tablet (this to prevent the enzymes from being disintegrated by gastric secretions) that contained 100 mg of rutoside, and 48 mg of trypsin, 90 mg of bromelain. Rescue medication, where necessary, was given in the form of ketorolac tablets 10 mg.

The radiographs (orthopantomography or intraoral periapical radiographs) were taken to assess third molar impaction regarding difficulty using WAR (White, Amber, and Red) lines and the Parant scale. In this double-blinded prospective randomized clinical trial, the primary outcome variable was postoperative edema which was assessed by measurements made between the 6 reference points of the Tragus, corner of the mouth, chin, angle of mandible, external canthus, and nasal bone. The secondary outcome variables were mouth opening measured using a caliper and pain assessed using a visual analog scale. These parameters were recorded on the 1st, 3rd, and 7th postoperative days respectively.

3. Statistical Analysis

The collected results underwent normality tests such as the Kolmogorov-Smirnov and Shapiro Wilk's test and the results revealed that the data followed a parametric distribution. For parametric continuous data, a student t-test was performed and to define statistical significance, a significance level of $p < 0.05$ was chosen. Statistical software (SPSS v.17 software, IBM, Armonk, New York) was used to conduct all the analyses.

4. Results

Of a total sixty-eight patients in this study, five patients were later excluded as they were lost during follow-up. Hence, a total of 63 patients were included in the final analysis who had attended the follow-up visits. The mean patient age in groups I and II was 27 and 28 years respectively. Differences in demographic characteristics of the subjects and surgical procedure parameters among the study groups were not statistically significant.

The mean preoperative edema in both the control and test groups was comparable. The mean postoperative swelling on the 1st postoperative day in Group I was 54.7 mm and in Group II was 53.2 mm which was not statistically significant ($p = 0.34$). There was an increasing trend in facial swelling till the 3rd postoperative day, which decreased on the 7th postoperative day. The highest was observed in both the groups on the third postoperative day and least on the 7th postoperative day. Similarly, the difference between mean facial swellings was highest on the 3rd postoperative day. The mean swelling measures were found to be non-significant when compared between the two groups but a significant difference in mean facial swelling was observed on day 1, day 3, and day 7 compared to the preoperative day within the group ($p<0.001$) [Table 1].

Table 1. Mean of Four Distances (mm) measured over the Swelling using Tape Measure Method among the treatment groups				
Treatment groups	Day 0	Day 1	Day 3	Day 7
Serratiopeptidase n=31	52.9 ± 3.4	54.7 ± 3.4*	55.1 ± 3.0*	52.9 ± 3.4 # ^
Enzyme combination n=32	51.5 ± 3.2	53.2 ± 3.2*	54.2 ± 3.3* #	51.9 ± 3.3* # ^
p-value	0.40	0.34	0.17	0.20
Data expressed as mean ± SD, p-value within group: * p<0.001 compared to Day 1; # p<0.001 compared to Day 3; ^ p<0.001 compared to Day 7 [Student's 't' test]				

The mean postoperative pain score immediately after surgery was 8.3 ± 0.9 in group I and 8.2 ± 0.5 in group II. The mean pain score was highest on the day of surgery, which progressively decreased by the 1st, 3rd, and 7th postoperative days. The mean pain scores were non-significant when compared between the groups but were significant when compared within the groups (p<0.001) [Table 2].

Table 2. Mean Pain Score Measured Using Visual Analog Scale among the treatment groups				
Treatment groups	Day 0	Day 1	Day 3	Day 7
Serratiopeptidase n=31	8.3 ± 0.9	7.2 ± 0.8*	5.5 ± 1.6* #	2.4 ± 1.7* # ^
Enzyme combination n=32	8.2 ± 0.5	7.3 ± 0.9*	5.4 ± 1.2* #	2.4 ± 1.1* # ^
p-value	0.69	0.83	0.88	0.99
Data expressed as mean ± SD, p-value within group: * p<0.001 compared to Day 1; # p<0.001 compared to Day 3; ^ p<0.001 compared to Day 7 [Student's 't' test]				

Analysis of the data showed that the mean baseline measure of the inter-incisal distance was 44 ± 3.7 mm and 43.4 ± 8.2 mm in groups I and II respectively. In both the groups, trismus was greater on day one after surgery and resolved during the follow-up periods. There was no significant difference between the Serrapeptase group and the Enzyme combination group in the mean maximal interincisal distance during the follow-up period [Table 3]. Data regarding the use of rescue medication in the treatment group is presented in Figure 1. Of the 31 patients recruited in group I, only 3 (14.3%) required rescue medication (ketorolac) as compared to 6 out of 32 patients (18.8%) in group II. As per the Parant scale for difficulty grading for third molar removal, the majority of the cases were classified as grade 3 difficulty (76.9%).

Table 3. Mean of Mouth Opening (mm) Measured Using Calipers among the treatment groups				
Treatment groups	Day 0	Day 1	Day 3	Day 7
Serratiopeptidase n=31	44.4 ± 3.7	23.1 ± 6.7	29 ± 7.3	35.8 ± 8.2
Enzyme combination n=32	43.4 ± 8.2	21 ± 5.7	24.4 ± 8.2	31.3 ± 9.4
p-value	0.62	0.29	0.08	0.12
Data expressed as mean ± SD [Student's 't' test]				

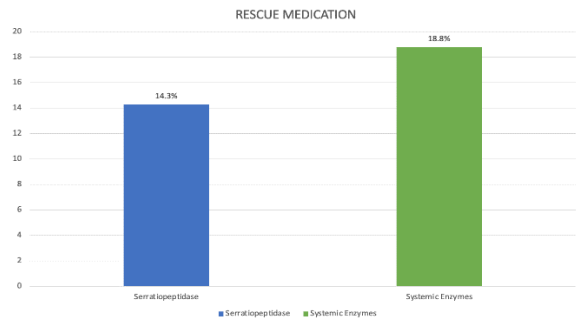

Figure 1: Requirement for Rescue Medications among the treatment groups

5. Discussion

The current study compared the efficacy of an enzyme combination (Trypsin, Bromelain, and Rutoside) with serratiopeptidase on the postoperative sequelae after surgical removal of impacted mandibular third molars. Gender, surgical experience, age, and operative time are all risk factors for pain, trismus, and edema after third molar surgery [9].

Mild bleeding, edema, stiffness, and pain are all common physiologic reactions following the surgical removal of an impacted third molar. All of these are perceived as unpleasant by the patient and should be avoided to the greatest extent possible. The postoperative pain begins when the effects of the local anesthesia wear off and peaks during the first twelve hours after surgery [10]. Third molar surgery is known to cause post-surgical edema or swelling. The swelling typically peaks by the end of the second postoperative day and resolves by the fifth to the seventh day. Postoperative events (pain, trismus, swelling) are usually treated with pharmacological drugs and/or other accessory methods such as the use of drains, ice packs, and laser therapy.

Facial swelling was evaluated by measurements made between 6 known standard reference points: a) Tragus, b) corner of the mouth, c) chin, d) angle of mandible, e) external canthus, and f) bridge of nasal bone. The horizontal measurements are the distances between the outer corner of the mouth and the tragus, as well as the distance from the tragus to the chin region. The distance between the outer canthus of the eye and the angle of the mandible, as well as the tip of the nasal bone to the angle of the mandible, is measured vertically. The facial measurement is determined by the arithmetic mean of the measurements. In our study, post-surgical pain

felt by the patients was higher on the day of surgery, attaining peak levels at 3-5 hours after the surgery which is in accordance with other studies [9-11]. The pain scores were also found to be non-significant.

Proteolytic enzymes are enzymes that are naturally produced by the human body and other living organisms to aid in the performance of essential functions. When these enzymes are supplemented, they have been shown to have an anti-inflammatory effect on body tissues. A study that compared the enzyme complex with placebo for edema control in orthognathic surgery concluded that usage of the enzymes statistically decreased post-op edema precluding long-term corticosteroid use [5].

Chopra et al [12], in their study, showed that when compared to betamethasone and ibuprofen, serratiopeptidase had no significant analgesic or anti-inflammatory effect, but it was better than placebo. Al-Khateeb and Nusair et al [13], compared serrapeptase and placebo in the reduction of post-op edema and stated that serrapeptase significantly decreases swelling and pain in all subjects, but there was no significant difference in mean maximal interincisal distance between the serrapeptase and placebo groups. Tamimi et al., have also reported recently that serratiopeptidase resulted in a better improvement in the inflammation compared to placebo [14].

Another study that compared bromelain with placebo found that there were no statistically significant differences between the groups, but the patients belonging to the bromelain group exhibited improved mouth opening and lesser inflammation [15]. In 2014, Majid and Al-Mashhadani compared bromelain to diclofenac and found that bromelain improves the quality of life following surgical removal of third molars. The investigators used 250 mg of bromelain four times a day in their study [16].

In the current study comparing the enzyme complex and serratiopeptidase, we found that the size of the swelling was almost equal in both groups and was not very significant. The results revealed that there was no significant difference in mouth opening reduction (trismus) between the study groups. This suggests that the enzyme complex is unlikely to be a powerful anti-inflammatory agent. A study reported that administering diclofenac potassium and dexamethasone did not reduce postoperative trismus [17].

Cysteine proteases (Bromelain, trypsin) and serine combinations are significant as they have different substrate specificities and a variety of effects, such as anti-edematous effects and effects on anti-proteinases and **α**2 macroglobulin, which are thought to contribute to their clinical efficacy [18-23]. The regulation of adhesion molecules, activation of fibrinolysis, the release of inflammatory mediators, and the breaking up of detritus contribute to better healing with systemic enzyme therapy. Enzymes also decrease immune complexes, which are involved in the pathophysiology of inflammatory rheumatic illnesses [24-29].

6. Conclusion

Within the limitations of our study, it can be concluded that there was no significant difference in the effect of the enzyme combination of rutoside, trypsin, and bromelain when compared to serratiopeptidase for the reduction of postoperative edema, pain, and trismus following lower third molar surgery. Future studies should be done with a higher dosage of the enzymes on a larger sample size to assess the efficacy of these drugs.

7. Conflict of Interest

The authors declare no conflicts of interest

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