

Evaluation of Some Properties of Prepared Indomethacin Mouth Wash and Its Effect on Gingival Index Score

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الخلاصة

الأهداف: تهدف الدراسة إلى تحضير غسول الاندوميثاسين الفموية ودراسة بعض خصائصها (الأشعة تحت الحمراء، قياس الباهة الحمضية، الكثافة، اللزوجة، الشد السطحي) وتقييم تأثيرها على مؤشر التهاب اللثة، ثم تحضير غسول الاندوميثاسين الفموية بتركيز (50) ملغ لكل (100) مل من الماء المقطر من مسحوق الاندوميثاسين النقي. **المواد وطرائق العمل:** جرت الدراسة على (40) شخصا تتراوح أعمارهم بين (20-30) سنة، قسموا إلى أربع مجاميع، كل مجموعة تضم عشرة أفراد، المجاميع الثلاثة الأولى مصابين بالتهاب اللثة، تم قياس مؤشر التهاب اللثة لهم قبل وبعد العلاج، حيث تضمن العلاج معالجة ميكانيكية بواسطة تنظيف وتلميع الأسنان في عيادة طب الأسنان، كما وتم استخدام علاج دوائي، حيث أعطيت للمجموعة الأولى غسول الاندوميثاسين الفموية، والمجموعة الثانية أعطيت غسول الكلورهيكسدين الفموية، في حين أعطيت المجموعة الثالثة الماء المقطر غسول فموي ثلاث مرات يوميا لمدة ثلاثة أيام، أما المجموعة الرابعة فقد تضمنت عشرة متبرعين لا يعانون من التهاب اللثة، تم تحليل البيانات باستخدام البرامج الإحصائية: اختبار (T) ، تحليل التباين، واختبار دنكن لتحليل البيانات. **النتائج:** أظهرت النتائج أن مؤشر التهاب اللثة انخفض معنويا في المجموعتين الأولى والثانية مقارنة مع قبل العلاج وبعده لنفس المجموعة، بينما في المجموعة الثالثة لا يوجد اختلاف معنوي قبل وبعد العلاج. **الاستنتاجات:** إن غسول الاندوميثاسين الفموية أظهرت تأثيرا مضادا لالتهاب اللثة وانخفاض في مستوى مؤشر التهاب اللثة.

ABSTRACT

Aims: to prepare Indomethacin mouth wash and study some of its properties (IR, pH, Density, Viscosity, Surface tension). Also, the effect on gingival index score were evaluated. Indomethacin mouth wash was prepared in a concentration of (0.05%) distil water from pure Indomethacin powder. **Methods:** The study was carried out on 40 subjects aged (20-30) years old. They were divided into four groups ten for each, three groups complaining of gingivitis. The gingival index score for all were measured before and after treatment, treatment involved mechanical treatment by scaling and polishing at dental clinic and drug treatment in which the first group received Indomethacin mouth wash, second group received Chlorhexidine mouth wash and third group received distal water as a mouth wash three times daily for three days, while the fourth group consisted of 10 volunteer subjects without gingivitis. Data were analyzed using paired t-test, ANOVA test and Duncan's Multiple analysis range test. **Results:** The results showed that the gingival index score levels were decreased significantly in all treatment groups between pre and post treatment at the same group. **Conclusions:** Indomethacin mouth wash have anti inflammatory effect that can decrease gingival index level. **Key words:** Indomethacin, Mouth Wash, Infrared spectroscopy.

Al-Nori MK, Taqa GA, Delemi ZH, Evaluation of Some Properties of Prepared Indomethacin Mouth Wash and Its Effect on Gingival Index Score. *Al-Rafidain Dent J.* 2014; 14(2):259-265.

Received: 6/5/2013 **Sent to Referees:** 8/5/2013 **Accepted for Publication:** 23/6/2013

INTRODUCTION

Gingivitis: Inflammation of the gums, characterized by pain, redness, swelling, and tendency to bleed. The gingival tissues are normally light pink, although the color may be related to the complexion of the person, the thickness of the tissue, and the

degree of keratinization. ⁽¹⁾ The accumulation of microorganisms on the tooth surface along the gingival margin can alter the structure and function of the gingiva, inducing an oral inflammatory reaction; clinically, this is known as gingivitis. ⁽²⁾

Indomethacin: Humans usually use

non steroidal anti-inflammatory drugs NSAIDs in various forms for more than 3500 years. Despite this long history, the mechanisms of how NSAIDs achieve their actions are still not completely unraveled. (3) Indomethacin introduced in 1963, is an indole derivative, it has anti-inflammatory, analgesic, and antipyretic activity, its non selective COX inhibitor. (4) Indomethacin is indicated for use in rheumatic conditions and is particularly popular for gout and ankylosing spondylitis in addition, it has been used to treat patent ductus arteriosus. (5) An ophthalmic preparation seems to be efficacious for conjunctival inflammation and to reduce pain after traumatic corneal abrasion. Oral administration of Indomethacin produces serious gastrointestinal adverse effects upon chronic administration. (6) The side effects of Indomethacin may include abdominal pain, diarrhea, gastrointestinal hemorrhage, and pancreatitis. Headache was reported in 15-25% of patients, associated with confusion and depression. Rarely psychosis with hallucination has been reported. Hematologic

reactions including thrombocytopenia and aplastic anemia. Hyperkalemia has also been reported and is related to inhibition of the synthesis of prostaglandins in the kidney. (7) Therefore, an alternative route is required to eliminate these oral adverse effects. Transdermal route and topical mouth wash has been known to eliminate oral adverse effects. (8) The aims of this study were to prepare of Indomethacin mouth wash, study some of its physicochemical properties (IR, pH, Density, Viscosity, Surface tension) and also to evaluate its effect on gingival index score.

MATERIALS AND METHODS

Preparation of Indomethacin solution

Indomethacin a weak acid is insoluble in water, it should be converted in to the soluble pharmaceutical form, sodium Indomethacin, by adding the equivalent amount of Na₂CO₃ (sodium carbonate) according to the molecular weight of them that show below: (9)

Material	Molecular weight
Indomethacin	357.79
sodium carbonate	106

Preparation of Indomethacin mouth wash

Indomethacin mouth wash was prepared by mixing (50)mg from Indomethacin powder (Ninawa Drug Industry) in (100)ml distilled water and adding (0.0148)g of sodium carbonate to give a final concentration of (0.05%) Indomethacin mouth wash. Magnetic stirrer was used to prepare a homogenous clear solution of sodium Indomethacin. The Indomethacin mouth wash was kept in a glass, amber color containers and store at room temperature. (10) Indomethacin mouth wash prepared freshly and given to the patients.

Infrared Spectroscopy (FTIR-Fourier transmittance infrared)

This test measured the vibration of bonding atoms, the infrared spectra recorded for Indomethacin powder and for the precipitate of sodium Indomethacin was examined by using Bruker Tensor 27 IR spectrophotometer (Germany) in the

region (500-4000 cm) using KBr disc. This measurement was carried out in University of Mosul, College of Education, Iraq. Sodium Indomethacin powder was obtained from drying of prepared Sodium Indomethacin solution at room temperature.

pH

Indomethacin and Chlorhexidine (Claridine) mouth wash pH was determined by putting the electrode of pH meter (Ecoscan/Singapore) inside the tube which contains few milliliters of the tested samples. The determination of pH value in Indomethacin mouth wash was done immediately after preparation, after three, six and nine months. Along these time intervals, the solution is clear, homogenous, and has the same taste and color density.

Density

The determination of density of the above samples was done by measuring its

mass per unit volume, in the metric system, density has units of kg/L or gm/ml. Water at (4⁰C) has density of exactly 1.00 gm/ml. Volumetric flasks were used in determination of density.⁽¹¹⁾

Viscosity

Determination of viscosity of Indomethacin and Chlorhexidine mouth wash was done by using Ostwald viscometer (England) which is a simple device and accurate for measuring the viscosity of liquid.⁽¹²⁾

Surface tension

The determination of surface tension of the above samples was done by the most accurate method which consisted of measuring the height to which the tested sample rises in a capillary tube.⁽¹³⁾ The surface tension of a liquid decreases as the temperature rises, the unit of surface tension is Dyn/cm or Newton/meter.

Effect of Indomethacin mouth wash on gingival index score

In this study, we took four groups, three groups of gingivitis patients (medically fit, aged 20-30 years, non smoker and non drinker) of subjects were participated, each group consist of 10 patients. The gingival index score for all was measured before and after treatment, at the same time, treatment involved mechanical treatment by scaling and polishing at dental clinic and drug treatment in which the first group prescribe Indomethacin mouth wash, the second group prescribe Chlorhexidine mouth wash and the third group prescribe placebo mouth wash (distil water) every 8 hours for three days. The forth group consisted of 10 volunteer subjects with good oral hygiene without gingivitis. Gingival index score: According to the

Gingival index, severity of gingivitis is assessed separately at the four smooth surfaces of the teeth, where ever the gingival margin looked inflamed it is massage with the side of the periodontal probe, if this message does not result in bleeding the unit score 1, if bleeding occurs, score 2 is giving, ulceration and spontaneous bleeding score 3.⁽¹⁴⁾

Statistical analysis

Data were analyzed using paired t-test, ANOVA test and Duncan's Multiple analysis range test, the level of significance was at $p < 0.05$.⁽¹⁵⁾

RESULTS

The vibrational response showed in Figure (1) of pure Indomethacin powder when passed via an infrared beam, while Figure (2) represents the vibrational response of precipitate of Sodium Indomethacin, the results in Figure (1) and (2) showed there are identical diagram for Indomethacin powder and for Precipitate of Sodium Indomethacin, this means that they have the same chemical structure. Evaluation of the pH of Indomethacin mouth wash was done immediately after preparation, after three, six and nine months showed (7.51, 7.55, 7.45, 7.21) respectively, for Chlorhexidine the pH results was (6.9, 6.88, 6.80, 6.60) respectively and for Distilled water the pH results was (7.40, 7.42, 7.38, 7.02) respectively, Table (1). The Density of Indomethacin, Chlorhexidine and Distilled water mouth washes was measured and the results (0.991, 1.029, 1) g/cm³ respectively, the Viscosity for them (0.991, 1.029, 1) poise. respectively, and the Surface tension for them (65.55, 31.76, 70.56) dyn/cm respectively. Table (2), Table (3), Table (4).

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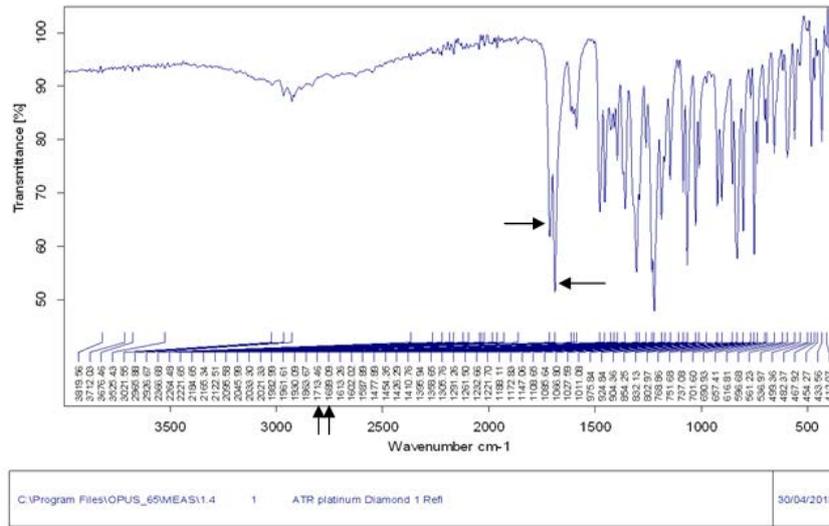


Figure (1): Pure Indomethacin powder measurement by FTIR (Infra-red) spectroscopy.

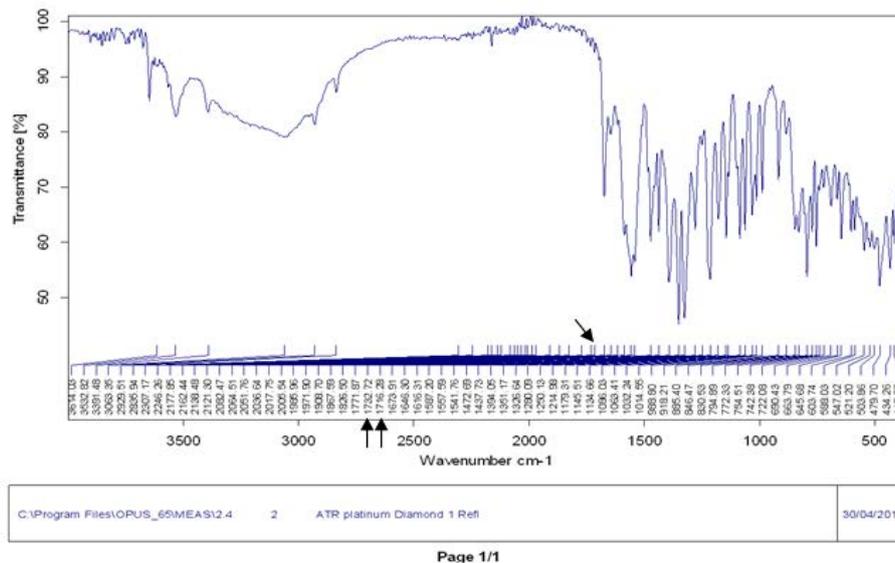


Figure (2): Precipitate of sodium Indomethacin powder measurement by FTIR (Infra-red) spectroscopy.

Table (1): pH of Indomethacin, Chlorhexidine and Distilled water mouth washes.

Material	pH at once	pH after 3 months	pH after 6 months	pH after 9 months
Indomethacin mouth wash	7.51	7.55	7.45	7.21
Chlorhexidine mouth wash	6.9	6.88	6.80	6.60
Distilled water	7.4	7.42	7.38	7.02

Table (2): Density of Indomethacin, Chlorhexidine and Distilled water mouth washes.

Material	Density g/cm ³
Indomethacin mouth wash	0.991
Chlorhexidine mouth wash	1.029
Distilled water	1

Table (3): Viscosity of Indomethacin, Chlorhexidine and Distilled water mouth washes.

Material	Viscosity poise
Indomethacin mouth wash	0.00873
Chlorhexidine mouth wash	0.003286
Distilled water	0.008937

Table (4): Surface tension of Indomethacin, Chlorhexidine and Distilled water mouth washes.

Material	Surface tension dyn/cm
Indomethacin mouth wash	65.55
Chlorhexidine mouth wash	31.76
Distilled water	70.56

Effect of Indomethacin mouth wash on gingival index score: The results showed that the gingival index score levels were decreased significantly in the Indomethacin, Chlorhexidine and Distilled water groups between pre treatment (1.76+0.38), (1.4+0.16), (1.48+0.30) respectively and post treatment (1.12+0.30),

(0.75+0.37), (1.12+0.34) respectively at the same group. Table (5), Figure (3). According to the Table (7), there were significance difference between post treatment effect of Indomethacin and Chlorhexidine, and that Chlorhexidine had lead to a better control of gingivitis score. Table (6), Table (7).

Table (5): Comparison between gingival index score before and after treatment.

Group	Gingival index	No.	Mean	+ SD	t-value	df	p-value
Indomethacinmouth wash	Before treatment	10	1.67	0.380	4.271	9	0.002**
	After treatment	10	1.12	0.304			
Chlorhexidine mouth wash	Before treatment	10	1.40	0.160	5.021	9	0.001**
	After treatment	10	0.75	0.370			
Distilled water mouth wash	Before treatment	10	1.48	0.300	2.455	9	0.036*
	After treatment	10	1.12	0.345			
Control	Healthy volunteers	10	0.51	0.177	-	-	-

** indicated significant difference at $p < 0.01$.

* indicated significant difference at $p < 0.05$.

Table (6): One way ANOVA of gingival index score among groups after treatment.

	Sum of Squares	df	Mean Square	F-value	p-value
Between Groups	2.690	3	9.410	9.410	0.000**
Within Groups	3.430	36			
Total	6.120	39			

** indicated significant difference at $p < 0.01$.

Table (7): Comparison between gingival index score among groups after treatment.

Group	N	Subset for alpha=0.05	
		1	2
control	10	0.516 (A)	
Distill water	10	0.757 (A)	
Chlorhexidine	10		1.124 (B)
Indomethacin	10		1.128 (B)

*The same letter indicates no statistical significant difference.

DISCUSSION

FTIR spectroscopy: In Figure (1) we see the peak characteristic for the carboxylic acid dimer (1713cm⁻¹) of amorphous Indomethacin, and for sodium Indomethacin in Figure (2) dimer peak has been eliminated. Also, absent in the shoulder at (1732cm⁻¹) that occurs because of the vibration of the non-hydrogen bonded carbonyl on the end of the chain. Indeed, even in the anhydrous and trihydrate crystal form sodium Indomethacin also lacked any peaks associated with dimer formation, as found in various forms of Indomethacin. The ionization of the carboxyl group and its electrostatic interaction with sodium ion would be expected to interfere with such dimer formation.⁽¹⁶⁾

The result obtained from the present study about the effect of Indomethacin mouth wash to reduce gingival index score and improve the gingival tissue condition may be returned to the mechanism of action of Indomethacin as a potent nonselective COX inhibitor and may inhibit phospholipase A and C, reduce neutrophil migration and decrease T cell and B cell proliferation.⁽⁴⁾ Gingival inflammation is reduced after administration of Indomethacin oral rinse.^(5, 17) These findings suggest that preparation of sodium Indomethacin mouth wash and use it in a concentration of 50mg/100ml is a new approach to attenuate topical inflammatory condition, and it has an important impact in order to create an effective and inexpensive oral health intervention for low socioeconomic communities, in this study the mechanical treatment with Indomethacin mouth wash, Chlorhexidine mouth wash reduce the gingival index score in all treatment groups and reduce the gingivitis. After preparation of Indomethacin mouth wash and study some of its properties, the pH of prepared mouth wash was nearby to Chlorhexidine and still the pH almost constant even with long period of preparation, this indicates that Indomethacin mouth wash was pH stable even in long period. These results are in agreement with several studies that have shown that the chewing stick is as or more effective than the toothbrush in reducing plaque and gingivitis.⁽¹⁸⁾ Moreover, the results of

another study confirm that Salvadorapersicamiswak use has a significant effect on dental plaque. Therefore, Salvadorapersicamiswak can be used as a dental hygiene method in combination with interproximal cleaning aides.⁽¹⁹⁾ Also, the results of this study agreed with previous study that found 0.19% Azadirachtaindica (neem) has significant anti-inflammatory property, thus it can be used as adjunct to mechanical therapy for treating plaque induced gingivitis.⁽²⁰⁾ Another study suggest that the clinical benefits of anti-plaque, antigingivitis mouth rinses are similar to the benefits of oral prophylaxis and oral hygiene instructions at six month recall appointments.⁽²¹⁾ In the present study, the use of Chlorhexidine reduces gingival index score, this result agreement with previous study suggested that in gingivitis patients, Chlorhexidine mouth rinses together with oral hygiene versus placebo, or control mouth rinse provide significant reductions in plaque and gingivitis scores, but a significant increase in staining score.⁽²²⁾ The present study, found that Indomethacin mouth wash and Chlorhexidine mouth wash have the same efficacy as a mouth wash from study of their effect on gingivitis patient and the two mouth wash have the ability to reduce gingival index score.

CONCLUSION

Although only a small number of anti-inflammatory mouth wash agents are available for use in local conditions. The obtained results demonstrate that the topical Indomethacin mouth wash may provide clinicians with a new option in the battle against gingival inflammation.

REFERENCES

1. Delaney JE, Keels MA. Pediatric oral pathology. Soft tissue and periodontal conditions, *Pediatric Clin Nort Am*. 2000; 47(5):1125-1147.
2. Mariotti A. Dental plaque-induced gingival diseases. *Ann Periodontol*. 1999; 4:7-19.
3. Vane JR .The fight against rheumatism: from willow bark to COX-1 sparing drugs. *J Physiol Pharmacol*. 2000; 51:573-586.

4. Richard F, Luigi XC, Michelle AC. Lippincott's Illustrated Reviews. 4th edition. 2009; P:507.
5. Katzung- B G, Master SB, Trevor A J. Therapeutic and clinical pharmacology. 12ed. Basic concentration. Copyright. McGrawhill companies., California, 2012; P638.
6. Badawi AA, El-Laithy HM, El-Qidra RK, El-Mofty H, El-Dally M, Chitosan based nanocarriers for Indomethacin ocular delivery. *Arch Pharm Res*, 2008; 31:1040-1049.
7. Yagiela AG, Jone FG, Dowed FJ, Johnson BS, Mariotti AJ, Neidle EA. Pharmacology and therapeutics for dentistry. American Dental Association. New-York 6th ed. 2011; P: 78-161.
8. Baboota S, Shakeel F, Kohli K, Development and evaluation of once a day transdermal gel of diclofenac diethylamine. *Method Find Exp Clin Pharmacol*. 2006; 22(2): 14-109.
9. Basha S, Keane D, Morrissey A, Nolan K, Oelgemöller M, Tobin J. Studies on the Adsorption and Kinetics of Photo degradation of Pharmaceutical Compound, Indomethacin Using Novel Photo catalytic Adsorbents. 2010; 49: 11302-11309.
10. Lin SC, Jiunh L . Indomethacin, Anti-inflammatory effects of Indomethacin mouth rinse *CDJ*, 1983; 2(1): 33-36.
11. Skoog DA, West DM . Fundamentals of analytical chemistry, third ed. Holt, Rinehart and Winston, New York, 1976; P704-705.
12. Woodbury G .Physical chemistry, 1st ed. Br.oki, publishin company, 1997pp 325-326.
13. Danials F , Alberty RA . Physical chemistry, Second ed. John Wily and Sons, INC, New York, London, 1983; P: 597-599.
14. Loe H and Silness J . Periodontal disease in pregnancy 1, Prevalence and severity, *Acta Odonto logica Scandinavica* 1963; 21: 533-551.
15. Daniel WW. Biostatics basic concepts and methodology for health sciences. *Wiely J Son .INC* 9th edition 2010; P: 346.
16. Taylor LS and Zografi G . spectroscopic characterization of interactions between PVP and Indomethacin in amorphous molecular dispersion. *pharm. Res*. 1997; 14:1691-1698.
17. Katzung- B G., Master SB, Trevor A J. Therapeutic and clinical pharmacology. 11ed. Basic concentration. Copyright. McGrawhill companies., califonia, 2009; P: 277.
18. Al-Otaibi M., Al-Harthy M., Soder, B., Gustafsson, A., Angmar-Mansson B . Comparative effect of chewing sticks and tooth brushing on plaque removal and gingival health. *Oral Health and Preventive Dentistry*, 2003; 1: 301-307.
19. AbierSofrataa, Fernanda Britoa, Meshari Al-Otaibib, Anders Gustafssona. Short term clinical effect of active and inactive Salvadorapersicamiswak on dental plaque and gingivitis, *Journal of Ethnopharmacology*, 2011; 137:1130-1134.
20. Chatterjee A, Salujam M, Singh N , KandwalA. To evaluate the antigingivitis and antiplaque effect of an Azadirachtaindica (neem) mouth rinse on plaque induced gingivitis, *Journal of Indian Society of Periodontology*, 2011; 15 (4):401.
21. John C.Gunsolley . Clinical efficacy of antimicrobial mouth renses, *Journal of dentistry*, 2010; 38: 6-10.
22. Van Strydonck DA, Slot DE, Van der Velden U , Van der Weijden F. Effect of a chlorhexidine mouthrinse on plaque, gingival inflammation and staining in gingivitis patients: a systematic review. *J Clin Periodontol*, 2012; 39: 1042–1055.