RESEARCH ARTICLE

Evaluation of Drop Volume Consistency of Eyedrop Bottles for New Generic Prostaglandin Analogue, Latanost® in Glaucoma Treatment

Jessica S. C. Ng[1], Yi X. Tan[1], Nor A. A. Alwi[1], Kar M. Yee[1], Ahmad H. A. Rashid[2], Ka-Liong Tan[3], Chuei W. Leong[1]*

[1]*Formulation R&D Technologies, Duopharma Innovation Sdn. Bhd., 40150 Shah Alam, Selangor, Malaysia.*
[2]*Industrial Biotechnology Research Centre, SIRIM Berhad., 40700 Shah Alam, Selangor, Malaysia.*
[3]*Faculty of Medicine and Health Sciences, Universiti Sains Islam Malaysia (USIM), Persiaran Ilmu, Putra Nilai, 71800 Nilai, Negeri Sembilan, Malaysia*

Received: 06th January, 2022; Revised: 29th March, 2022; Accepted: 10th April, 2022; Available Online: 25th June, 2022

ABSTRACT

Difficulty in instilling eyedrop is a concern for it may cause a progression of blindness in glaucoma patients. Early depletion of eyedrop before the next scheduled medication refill is a common issue faced by patients, resulting in poorer compliance to the therapy. Hence, we hypothesize that the lack of drop volume consistency is due to the eye drop bottle-related mechanics during drop instillation. This study aims to select the best bottle designs by examining drop volume consistency during drop instillation. Ten eyedrop bottles of different volumes were filled with 2.5 mL purified water. Next, the drop uniformity test was performed by expelling 10 droplets by holding the bottle vertically and weighing with an analytical balance. Based on the outcome, bottle C, which demonstrated high drop consistency with low percentage standard deviation and near-linear regression, was chosen as the eyedrop container for Latanost® 0.005% w/v eye drops. Using bottle C, the drop volume of Latanost® was estimated at 25.30 ± 1.87 µL. A 2.5 mL latanoprost solution can yield approximately 99.36 ± 7.52 drops per bottle. Besides that, the design of the eyedrop nozzle could determine the consistency of medication delivery. This report could better inform prescribers and patients in predicting the course of treatment. The selection of the right design of container closure system with a consistent drop delivery could significantly address the issue of early depletion of eyedrop before the next scheduled medication refill and difficulty in instilling eyedrop in glaucoma patients. This helps to improve medical treatment compliance in glaucoma patients.

Keywords: Drop volume consistency, Glaucoma treatment, Medication adherence, Prostaglandin analog.

International Journal of Drug Delivery Technology (2022); DOI: 10.25258/ijddt.12.2.3

How to cite this article: Ng JSC, Tan YX, Ahmad Alwi NA, Yee KM, Abdul Rashid AH, Tan K-L, Leong CW. Evaluation of Drop Volume Consistency of Eyedrop Bottles for New Generic Prostaglandin Analogue, Latanost® in Glaucoma Treatment. International Journal of Drug Delivery Technology. 2022;12(2):476-480.

Source of support: Nil.

Conflict of interest: K. L. Tan underwent a 6 months industrial attachment with Duopharma Biotech Berhad under Ministry of Higher Education, Malaysia.

INTRODUCTION

Glaucoma is a progressive optic neuropathy that leads to the loss of retinal ganglion cells associated with the loss of the visual field.[1] It is the second leading cause of global blindness. However, with early diagnosis, the patients could manage their condition through medication and surgery.[2] The health burden of glaucoma, on the other hand, has been increasing continuously over the past 25 years.[3] In Malaysia, 6.6% of blindness was reported caused by glaucoma.[4] The disease burden is expected to increase in the future. The management of glaucoma could be achieved by reducing intraocular pressure by topical ocular application of antihypertensives, *i.e.*, prostaglandin analogs, β-blockers, adrenergic agonists, cholinergic agents, and osmotic agents.[5] Compared to the others, prostaglandin analogs are recommended as the first-line medication in glaucoma treatment in Malaysia.[6]

Latanoprost is a prostaglandin F2α analog which exerts agonist activity with improved selectivity towards prostaglandin F-receptor. Prostaglandin F2α is one of the prostaglandins that is naturally synthesized in the iris and ciliary body within the eye. It enhances the outflow of aqueous humor through the uveoscleral route and trabecular meshwork leading to reduced intraocular pressure.[7] Additionally, latanoprost is an esterified prodrug of prostaglandin F2α that undergoes rapid hydrolysis to form the active latanoprost acid. Within one to two hours post topical application, the concentration of the active drug

**Author for Correspondence: cwleong@duopharmabiotech.com*

reaches its peak in the aqueous humor, up to 15 to 30 ng/mL.[8] Hence, the recommended dosage is 1.5µg onto the affected eye once every evening.

In topical ophthalmic medications, multi-dosed eyedrop bottles are widely used for their cost-effectiveness. Approximately 25% of the patients encounter early eyedrop bottle exhaustion and thence adherence issue. Early eyedrop bottle exhaustion could be due to problems related to the bottle mechanics resulting in accidental large volumes of medication being administered. Several factors could affect dispensed droplet volume, *i.e.*, design of the eyedrop nozzle, surface tension, viscosity and density of the drug solution, handling angle of the container and squeezing force.[9–11] Whereby the strength of squeeze, handling of the eyedrop bottle, drop formation and the positioning of the bottle during application determine the final volume of the drop.[12] Furthermore, manufacturing and design issues could lead to the lack of uniformity of dispensing drops.[13,14] The wide variability in the drop volume is due to the lack of regulations.[13]

Varying latanoprost drug responses are due to varying drop volumes administered.[15] A smaller drop could reduce the therapeutic effect. Whereas an excessive amount could lead to higher systemic bioavailability through ocular absorption pathways, causing unwanted adverse effects.[16] Inadequate amount of latanoprost may jeopardize the patient's dose adherence. Inconsistent drop volume and a reduced number of doses per bottle may cause early bottle exhaustion, leading to poor medication compliance among patients, medication waste, and increased cost of treatment.[16,17] In the long run, the increasing cost of ophthalmology healthcare is becoming a concern. Glaucoma implies a significant financial impact on the patients as it requires long-term medications, surgery, consultations, and regular retinal examinations. The indirect costs also include caregivers for the visually impaired and with rehabilitation and disability to work.

Therefore, through the new generic latanoprost, Latanost®, researchers value the consistent dose delivery using efficient eyedrop bottles. The best eyedrop design must be selected and implemented to ensure cost-effectiveness for chronic glaucoma treatment. This study aims to assess the dropping consistency of eyedrop bottles from various manufacturers. The eyedrop bottle with a high dosing consistency will then be selected for the formulation of a generic latanoprost 0.005% (w/v) solution.

MATERIALS AND METHODS

Ten eyedrop bottles made by different manufacturers were purchased (Figure 1). The sample bottles represented a vast variety of regional brands and generic medications available in the market. All bottles were filled with 2.5 mL of water via pipette. The drop volume consistency was assessed by expelling 10 droplets by holding the bottle upright (90° angle) at room temperature. The drop volumes were weighed using an analytical balance (AB204S and XPR204, Mettler Toledo, US). The volume (µL) of the droplet was calculated from the drop weight (1g = 1 mL). The relative standard deviation (RSD) was calculated. Finally, a cumulative drop volume of 10 drops was plotted.

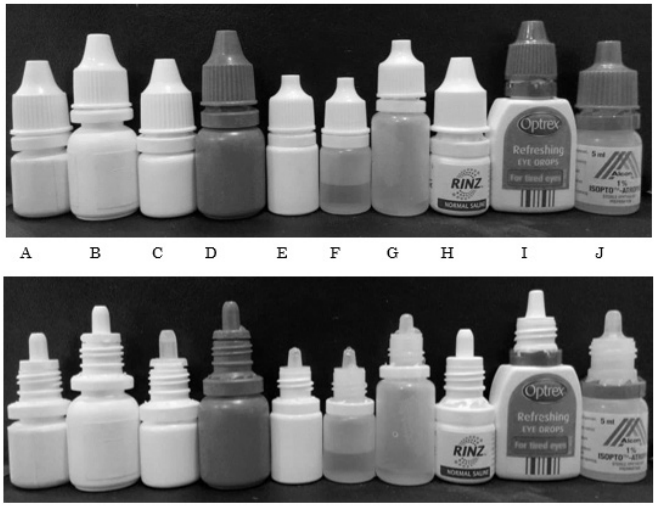
The bottle which demonstrated consistent dosing performance with low reflex sympathetic dystrophy (RSD) was selected as the container of choice for the formulation of latanoprost 0.005% (w/v) solution. Similarly, to compare our generic product to the branded formulation, drop consistency test was performed on the selected bottle filled with 2.5 mL latanoprost 0.005%(w/v) solution, Latanost®, and Xalatan®. The volume (µL) of the droplet was compared with the drop weight from density measurement.

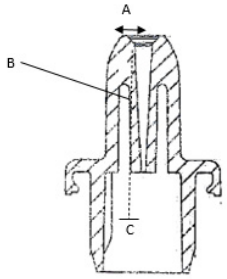
Statistical Analysis

Statistical analysis comprised of an analysis of variance (ANOVA), student t-test and regression linear analysis. ANOVA compared mean values for a two-way layout with bottles. Significant differences among the bottles were also obtained ($p < 0.05$). Additionally, a post-hoc comparison of means was performed by comparing the means between the positions of each bottle using the student t-test.

RESULTS

In this study, two parameters were evaluated to assess drop volume consistency. RSD (%) indicates the degree of variation from the mean of a pool of drop volume measurements. While the regression constant displays the consistency of drop volume from consecutive delivery from the eyedrop bottles. Using





A: Outer diameter of tip, B: Internal space volume, C: Inner aperture diameter

Figure 1: Tested bottles and the cross-sectional diagram of bottle C.

purified water in the sample bottles, the multi-dose bottles delivered single drop volumes in the range of 24.12 ± 5.95 to 50.93 ± 2.77 µL (Table 1). Based on the values, there was significant variation in the average drop volume expelled from the bottles ($p < 0.05$, ANOVA). When comparing eyedrop bottles of the same volume, significant differences were evident (Table 2), except for Bottle C and H. Bottle A and J exhibited the highest and lowest RSD%, respectively (Figure 2). According to the regression analysis of the cumulative drop volume of 10 droplets (Figure 3 and Table 3), bottle C and I yielded a regression constant (R^2) of close to 1.

Based on the findings, bottle C was chosen as a suitable container for Latanost® 0.005% (w/v) formulation due to its high consistency in drug delivery with low RSD%. Considering the volume of our marketed Latanost® eyedrop, which is 2.5 mL, a 5 mL bottle is more sustainable. Due to the higher viscosity of the drug solutions contributed by the addition of excipients, the weight of a drop of Latanost® is estimated at 0.0256 ± 0.0019 g, equivalent to 25.30 ± 1.87µL (density of Latanost® 0.005% (w/v) formulation = 1.01 ± 0.01 g/mL) (Figure 4). Figure 4 showed the Latanost® bottle is consistent and comparable to the innovator.

DISCUSSION

The increase in internal pressure when a patient squeezes the sides of the bottle leads to significant drop volume variability.

Table 1: List of investigated eyedrop bottles. volume of drops expelled from each bottle were expressed as mean ± SD and % relative standard deviation.

Bottle	*Source*	*Category*	*Labeled bottle volume (mL)*	*Drop volume expelled (µL)*	*%RSD*
A	Falcon Plastics	Empty bottle	5	33.61 ± 9.97	29.68
B	Falcon Plastics	Empty bottle	10	31.38 ± 7.87	25.08
C	Dr Pack	Empty bottle	5	41.10 ± 2.87	6.97
D	Dr Pack	Empty bottle	10	38.45 ± 5.03	13.07
E	Unknown	Empty bottle	5	24.12 ± 5.95	24.69
F	Unknown	Empty bottle	5	26.69 ± 4.51	16.88
G	Unknown	Empty bottle	10	25.32 ± 4.57	18.04
H	Rinz	Normal saline eye drop	5	41.91 ± 3.86	9.20
I	Optrex Refreshing Eye Drops for tired eyes	Lubricating eye drop	10	50.93 ± 2.77	5.44
J	Alcon	Anticholinergic eye drop	5	38.65 ± 2.06	5.34

Table 2: Differences in mean drop volume between bottles of the same volumes from different manufacturers as listed in Table 1. $p < 0.05$ indicates significant difference.

Bottle volume	*Bottles for comparison*	*Comparison*	*Results*	*Difference between mean drop volumes (µL)*	*p*
5 ml	A, C, E, F, H and J	A vs C	C > A	7.49	<0.001
		A vs E	A > E	9.49	<0.001
		A vs F	A > F	6.92	<0.001
		A vs H	H > A	8.30	<0.001
		A vs J	J > A	5.04	<0.001
		C vs E	C > E	16.98	<0.001
		C vs F	C > F	14.41	<0.001
		C vs H	H > C	0.81	0.18
		C vs J	C > J	2.45	0.001
		E vs F	F > E	2.57	0.03
		E vs H	H > E	17.80	<0.001
		E vs J	J > E	14.53	<0.001
		F vs H	H > F	15.22	<0.001
		F vs J	J > F	11.96	<0.001
		H vs J	H > J	3.27	<0.001
10 mL	B, D, G and I	B vs D	D > B	7.07	<0.001
		B vs G	B > G	6.06	<0.001
		B vs I	I > B	19.55	<0.001
		D vs G	D > G	13.13	<0.001
		D vs I	I > D	12.48	<0.001
		G vs I	I > G	25.61	<0.001

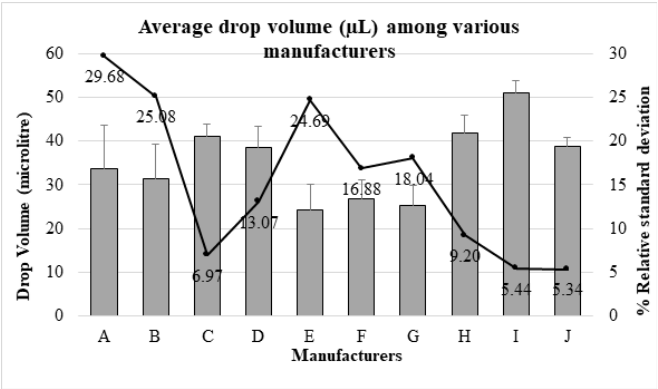


Figure 2: Bar chart showing the average drop volume of various bottles from all manufacturers with SD error bars (n = 3) with line graph depicting the percentage relative standard deviation.

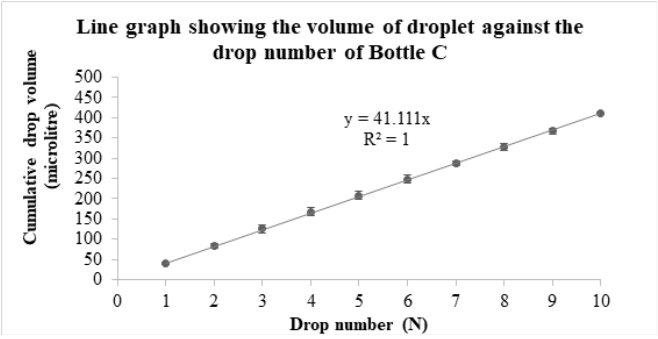


Figure 3: Line graph showing linear regression analysis of 10 droplet volume from Bottle C.

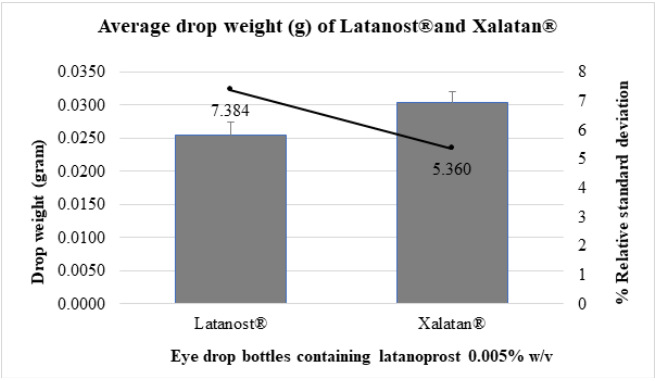


Figure 4: Bar chart showing the average drop weight of Latanost® and Xalatan® with SD error bars (n = 3) with line graph depicting the percentage relative standard deviation.

The internal pressure causes the liquid to flow from the inner aperture through the capillary of the nozzle to the outer tip forming a drop. Moreover, eyedrop volume is also directly dependent on the characteristics of ophthalmic solutions such as liquid surface tension and nozzle shapes, i.e., the diameter of the inner aperture and outer tip together with the internal space volume [18] (Figure 1). Hence, the differences in drop size and consistency were primarily attributed to the differences in the nozzle dimensions and designs. We identified that the formulation has a 1.01 ± 0.01 g/mL density, and it is interchangeable with water density estimated at 1.0 g/mL. However, the influences of the physical characteristics of the solutions (viscosity and surface tension) were eliminated in this study as the experiment was performed using only water.

Table 3: Regression constant of cumulative drop volume (µL) of 10 drops.

Bottle	*Regression constant*
A	0.9983
B	0.9988
C	0.9998
D	0.9991
E	0.9970
F	0.9990
G	0.9990
H	0.9996
I	0.9999
J	0.9996

Bottle C demonstrated high consistency in drug delivery and was chosen as the container for Latanost® 0.005% (w/v) formulation. With bottle C. It was estimated that a 2.5 mL of Latanost® solution would be able to yield approximately 99.36 ± 7.52 drops per bottle. This finding agrees with the number of drops per bottle of the innovator Xalatan® (with the same volume) which yielded between 87–92 drops.[14,19]

On the other hand, determining systemic bioequivalence is a mandatory registration requirement for generic products in Malaysia. Therefore, generic eyedrop should ensure comparability of the drop volume with the innovator product. It also ensures the amount of drug delivered into the eye is comparable to that of the innovator product. Once-daily application of latanoprost has been proven to sustain the reduction of intraocular pressure.[20] Persistent daily administration of glaucoma medications is necessary to maintain long-term therapeutic effects. Therefore, consistent uniformity with every dose administered to the eye during each application minimizes the variation in pharmacological effects.

In reference to the findings, the drop volume consistency is deemed essential for a generic medication in common clinical practice. Hence, choosing the right bottle design reduces the risk of sub-therapeutic dose administration or dose exceeding the prescribed dose. The bottle design could also guide prescribers and patients to predict the course of treatment better. It would prevent early exhaustion of medication and improve patient compliance with treatment.

Despite the higher price among latanoprost options, Xalatan® is prescribed as the standard glaucoma treatment in Malaysia. The previous report found the price ratio of Xalatan® to other generic latanoprost ranges from 0.6 to 3.5.[21] Pharmacoeconomic perspectives consider cost-effectiveness and patient affordability when considering generic brands. Hence, Latanost® is the more affordable option for the middle-income market.

This report is the first-ever generic ophthalmic medication product with the search to determine the best suitable bottles

to resolve user concerns. However, the notable limitation of the study was the employment of a single installation angle of 90º for testing. We understand it is unlikely that patients administer ophthalmic solutions at a strictly vertical position in practice. Hence, it is advisable to educate patients that a dispensing angle close to 90º is most efficient. A properly dispensed droplet volume leads to more consistent drug delivery.

CONCLUSION

This study demonstrated a lack of consistency in medication delivery bottles from various bottle manufacturers. The evaluation of drop volume consistency identifies manufacturer C as a suitable eyedrop bottle. Consistent drug dosing and pharmacoeconomic impact in glaucoma medications ensure the sustained treatment to the patients.

ACKNOWLEDGEMENT

We thank SIRIM Berhad for the work on collecting part of the drop volume data. This work was funded by Duopharma Biotech Berhad. Registration No: 200001021664 (524271-W). Professional medical English editing was done by *Proofreading By a UK PhD*. This study was supported by research innovation fund from Duopharma Biotech Berhad.

REFERENCES

1. Martucci A, Nucci C. Evidence on neuroprotective properties of coenzyme Q10 in the treatment of glaucoma. Neural Regen Res. 2019;14(2):197-200.
2. Patel A, Cholkar K, Agrahari V, Mitra AK. Ocular drug delivery systems: An overview. World J Pharmacol. 2013;2(2):47-64.
3. Wang W, He M, Li Z, Huang W. Epidemiological variations and trends in health burden of glaucoma worldwide. Acta ophthalmologica. 2019;97(3):e349-e55.
4. Chew FLM, Salowi MA, Mustari Z, Husni MA, Hussein E, Adnan TH, et al. Estimates of visual impairment and its causes from the National Eye Survey in Malaysia (NESII). PloS one. 2018;13(6):e0198799.
5. Beidoe G, Mousa SA. Current primary open-angle glaucoma treatments and future directions. Clinical ophthalmology. 2012;6:1699-707.
6. Jelinar M. Clinical Practice Guidelines Management of Glaucoma (Second Edition). Malaysian Health Technology Assessment Section, Medical Development Division, Ministry of Health Malaysia. 2017.
7. Alm A. Latanoprost in the treatment of glaucoma. Clinical ophthalmology. 2014;8:1967-85.
8. Sjoquist B, Stjernschantz J. Ocular and systemic pharmacokinetics of latanoprost in humans. Survey of ophthalmology. 2002;47 Suppl 1:S6-12.
9. Moore DB, Beck J, Kryscio RJ. An objective assessment of the variability in number of drops per bottle of glaucoma medication. BMC ophthalmology. 2017;17(1):78.
10. Moore DB, Hammer JD, Akhtari R, Beck J, Sanders S, Kryscio RJ. Squeeze Me if You Can: Variability in Force Requirements to Extract a Drop From Common Glaucoma Bottles. Journal of glaucoma. 2016;25(9):780-4.
11. Van Santvliet L, Ludwig A. Influence of the physico-chemical properties of ophthalmic viscolysers on the weight of drops dispensed from a flexible dropper bottle. European journal of pharmaceutical sciences : official journal of the European Federation for Pharmaceutical Sciences. 1999;7(4):339-45.
12. Fiscella R, Wilensky JT, Chiang TH, Walt JG. Efficiency of instillation methods for prostaglandin medications. J Ocul Pharmacol Ther. 2006;22(6):477-82.
13. German EJ, Hurst MA, Wood D. Reliability of drop size from multi-dose eye drop bottles: is it cause for concern? Eye. 1999;13 (Pt 1):93-100.
14. Queen JH, Feldman RM, Lee DA. Variation in Number of Doses, Bottle Volume, and Calculated Yearly Cost of Generic and Branded Latanoprost for Glaucoma. American journal of ophthalmology. 2016;163:70-4 e1.
15. German EJ, Hurst MA, Wood D. Eye drop container delivery: a source of response variation? Ophthalmic & physiological optics : the journal of the British College of Ophthalmic Opticians. 1997;17(3):196-204.
16. Davis SA, Sleath B, Carpenter DM, Blalock SJ, Muir KW, Budenz DL. Drop instillation and glaucoma. Curr Opin Ophthalmol. 2018;29(2):171-7.
17. Moore DB, Walton C, Moeller KL, Slabaugh MA, Mudumbai RC, Chen PP. Prevalence of self-reported early glaucoma eye drop bottle exhaustion and associated risk factors: a patient survey. BMC ophthalmology. 2014;14(1):79.
18. Yoshikawa K, Yamada H. Influence of container structures and content solutions on dispensing time of ophthalmic solutions. Clinical ophthalmology. 2010;4:481-6.
19. Kolko M, Koch Jensen P. The physical properties of generic latanoprost ophthalmic solutions are not identical. Acta ophthalmologica. 2017;95(4):370-3.
20. Alm A, Widengard I. Latanoprost: experience of 2-year treatment in Scandinavia. Acta ophthalmologica Scandinavica. 2000;78(1):71-6.
21. Zhao PY, Rahmathullah R, Stagg BC, Almobarak F, Edward DP, Robin AL, et al. A Worldwide Price Comparison of Glaucoma Medications, Laser Trabeculoplasty, and Trabeculectomy Surgery. JAMA ophthalmology. 2018;136(11):1271-9.