Table 4: Zone of inhibition values of formulation F1 to F6 and pure drug at the concentration of 2 µg/ml against *Staphylococcus aureus* and *Escherichia coli*

Microorganism	Concentration (mcg/ml)	Zone of Inhibition (mm)						
		Standard (Pure Drug)	F1	F2	F3	F4	F5	F6
Gram +ve *Staphylococcus aureus*	2 µg/ml	29.66	28	28.66	29	28.33	28	28.66
Gram -ve *Escherichia coli*	2 µg/ml	31.0	28.7	29.4	30.33	30.88	30.77	30.99

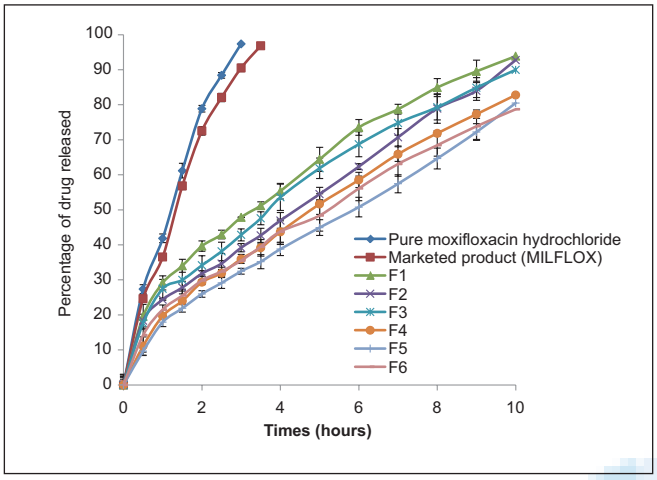

Figure 3: Comparative *in vitro* diffusion profile of pure drug, marketed product and F1 to F6 formulations

From the *in vitro* results it was observed that percentage release of the drug from the developed formulations F1 (93.86%), F2 (92.78%), F3 (89.97%), F4 (82.80%), F5 (80.49%), and F6 (78.71%) as shown in Figure 3. Formulation F6 showed more sustained release compared to other formulations. This could be the reason of higher concentration of Sodium alginate and HPMC K4M among the developed formulations. By observing the drug release profile it can be conclude that release is not stagnant even end of 10 hours. Formulation F6 showed highest zone of inhibition values against *S. aureus* (28.66 mm) and *E. coli* (30.99 mm), respectively, compared to other developed formulations. Hence, F6 formulation was taken for further study.

Antimicrobial efficacy study was performed on F6 formulation using Gram +ve *S. aureus* and Gram -ve *E. coli* organism. The zone of inhibition of F6 ophthalmic formulation found to be 28.66 and 30.99 mm, respectively, for Gram +ve *S. aureus* and Gram -ve *E. coli* organism. The results of antimicrobial activity are as shown in the Table 4. The study indicated moxifloxacin hydrochloride retained its antimicrobial activity when formulated as gel forming ophthalmic system against both selected *S. aureus* and *E. coli.*

Ocular irritation study was performed using healthy albino rabbits after getting prior permission from the institutional animal ethics committee. The eyes of each rabbits were examined at particular time interval after instillation of the optimized formulation (F6). There was no redness, continuous blinking, swelling or watering of eyes. No ocular damage or abnormal clinical signs to the cornea, iris or conjunctiva were visible. The result of ocular irritation studies indicates that formulations containing all ingredients are non-irritant to rabbit eye.

Accelerated stability studies were carried out at 40 ± 2°C at 75 ± 5 % RH for 1 month using stability chamber. The samples were analyzed periodically on every week, and found that there are no changes in visual appearance, clarity, pH, and gelation. Assay values after 1 month of storage are found almost same (deviating not more than one percent). Release profiles were similar to that of zero days.

CONCLUSION

Moxifloxacin hydrochloride, a broad spectrum antibacterial agent used in the treatment of ocular infections, was successfully formulated as *in situ* gel-forming eye drops using Sodium alginate as a gelling agent in combination with HPMC as a viscosity enhancing agent. Thus, the developed formulation is a viable alternative to conventional eye drops by virtue of its ability to enhance bioavailability through its longer precorneal residence time and ability to sustain drug release. Also important is the ease of administration afforded and decreased frequency of administration resulting in better patient acceptance.

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How to cite this article: Mandal S, Thimmasetty MK, Prabhushankar GL, Geetha MS. Formulation and evaluation of an in situ gel-forming ophthalmic formulation of moxifloxacin hydrochloride. Int J Pharma Investig 2012;2:78-82.

Source of Support: Nil. **Conflict of Interest:** None declared.

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