

Supplementary material to

## Enhanced anti-inflammatory and antibacterial efficacy of a novel gum ghatti-infused polymeric film for ocular drug delivery

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### Materials and methods

#### Physicochemical properties

Physicochemical properties such as weight uniformity, ocular composite thickness, surface pH, opacity, folding endurance, and blood biocompatibility were measured. The fabricated ocular composite was cut into a 1×1 cm<sup>2</sup> piece, weighed with a digital balance (Wensar, LPB21), and the weight was recorded. Five or six random portions of the film were selected, and the thickness was measured using a digital micrometer (Mitutoyo, Japan). The 1×1 cm<sup>2</sup> films were immersed in a petri dish containing 5 ml of double-distilled water. After 10 minutes, the water's pH was measured with a pH meter (L1617, Elico, Mumbai, India). To assess opacity, the film was cut into 40×50 mm pieces, and absorbance was recorded at 600 nm using a UV-Visible spectrophotometer (Shimadzu, Japan). Opacity was calculated using article Equation (1). Folding endurance was tested until a 2×2 cm<sup>2</sup> film was broken. Blood containing sodium citrate was used to evaluate hemolysis. In brief, 4 mL of sodium citrate blood was mixed with 5 mL of normal saline. Negative control consisted of 0.5 mL citrated blood with 9.5 mL normal saline; positive control contained 0.5 mL 0.1 M HCl, 0.5 mL blood, and 9 mL normal saline; and the test samples were prepared from previously collected lechate (incubated for 1 hour at 37 °C; 9 mL normal saline with 0.5 mL citrated blood and 0.5 mL lechate). All samples were incubated again for 1 hour at 37 °C, then centrifuged at 300 rpm for 10 minutes. The supernatant's absorbance was measured spectrophotometrically at 545 nm, and coagulation was calculated using article Equation (2)[1,2]. All physicochemical properties were expressed as mean ± SD (*n* = 6).

#### Drug release profile

Diffusion of the drug was assessed through a Franz diffusion cell. 12 ml of artificial tear fluid (ATF, composition: NaCl 6.8 g, NaHCO<sub>3</sub> 0.22 g, CaCl<sub>2</sub> · 2H<sub>2</sub>O 0.08 g, KCl 1.4 g added to deionised water and volume made up to 1000 mL) [3]. The donor and receiver compartment of the Franz cell was separated by a semipermeable dialysis membrane (MW: 60 kDa). A film (10 mm in diameter) was placed over the pre-hydrated dialysis membrane. Receptor compartment fluid temperature was maintained at 37 °C throughout the experiment. Stirring speed was maintained at 100 rpm with the help of a magnetic stirrer. The area of diffusion was 0.64 cm<sup>2</sup>. At a predetermined interval, 1 mL of the sample was removed and replaced with fresh ATF. Presence of the drug in the sample was measured through a UV-vis spectrophotometer (UV-1900i,

Shimadzu Corporation, Japan) at 289 nm. Different mathematical models like 1<sup>st</sup> order, Equation (S1); Higuchi, Equation (S2); Korsmeyer-Peppas, Equation (S3) and Peppas-Shalin, Equation (S4) were fitted to the observed data.

$$\frac{M_{\infty}}{M_i} = m = 100(1 - e^{-K_1 t}) \quad (S1)$$

where  $M_{\infty}$  is amount of drug at the equilibrium state,  $M_i$  is amount of drug released over time  $t$ ,  $m$  is fraction of the solute released,  $K_1$  is release rate constant and  $t$  = time.

$$\frac{M_{\infty}}{M_i} = m = k t^{0.5} \quad (S2)$$

where  $k$  is release rate constant.

$$\frac{M_{\infty}}{M_i} = m = k t^n \quad (S3)$$

where  $n$  is the diffusion exponent,

$$\frac{M_t}{M_{\infty}} = k_1 t^m + k_2 t^{2m} \quad (S4)$$

where  $k_1$ ,  $k_2$  and  $m$  are constants,  $k_1 t^m$  is Fickian diffusional contribution and  $k_2 t^{2m}$  is case-II relaxational contribution

## References

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