

Chloroquine Inhibitor

Catalog No:	NBP2-29386
Content:	200 mg
Storage:	The solid powder is stable in the desiccators at room temperature for 1 year. Water-reconstituted chloroquine solution is stable for up to 1 month at 4°C.
Species Reactivity:	N/A
Form:	Yellow Solid (Powder)
Inhibitor Mechanism:	Endosomal toll-like receptor inhibitor (antagonist); Inhibitor of endosomal acidification on which functional activity of endosomal TLRs (particularly TLR9 and TLR3) is dependent.

Background

Chloroquine is a weak base which can partition into acidic vesicles such as endosomes and lysosomes, resulting in inhibition of endosomal acidification and lysosomal enzyme activity. Because acidic pH of endosomes is a prerequisite of endosomal TLR activation, chloroquine can serve as an antagonist for endosomal TLRs. Chloroquine and its analog quinacrine are also known to act as therapeutic agents for autoimmune diseases such as rheumatoid arthritis and systemic lupus erythematosus, of which therapeutic activity is due to suppression of TLR9 activity as shown by researchers.

Solubility

Deionized water

Usage:

Product Handling Protocol

1. To make 100 mM stock solution, dissolve 200 mg chloroquine in 3.9 ml water by gentle vortex.
2. Filter sterilize through a 0.22 μ m filter.
3. Store at 4°C (Note: Chloroquine solution is light sensitive).
4. For TLR signaling inhibition study, perform a pilot inhibitory test with the different concentrations of chloroquine ranging from 1 to 10 mM to optimize your experiments.

Note: See our validation tests using the NBP2-26280 (chloroquine sensitive) and NBP2-26274 (chloroquine insensitive) cell lines as shown in Figures 1 and 2.

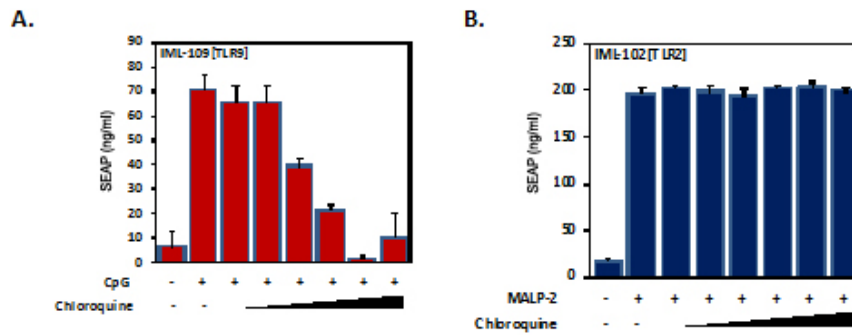


Figure 1. Evaluation of inhibitory activity of chloroquine. TLR9/NF- κ B/SEAPorter™ HEK 293 (NBP2-26280) and TLR2/NF- κ B/SEAPorter™ HEK 293 (NBP2-26274) cells were plated in 96-well plates at 5×10^4 cells/well for 16 h. Cells were preincubated with different concentrations of chloroquine (0.05, 0.1, 0.5, 1, 5 and 10 μ M) for 30 min. Cells were then stimulated with 10 μ g/ml CpG (NBP2-26232) [A] or 20 ng/ml MALP-2 (NBP2-26219) [B] for 24 h. Secreted alkaline phosphatase (SEAP) was analyzed using SEAPorter™ Assay Kit (NBP2-25285).

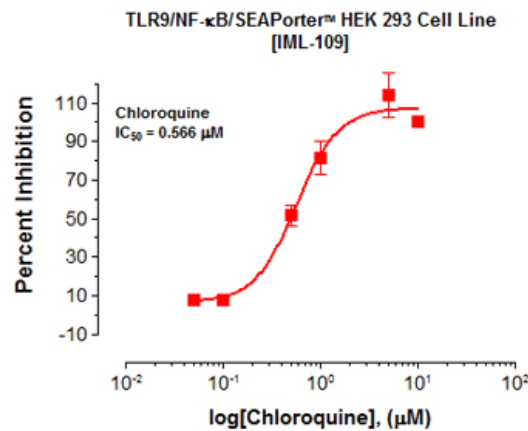


Figure 2. IC_{50} evaluation of chloroquine. TLR9/NF- κ B/SEAPorter™ HEK 293 (NBP2-26280) cells were plated in 96-well plates at 5×10^4 cells/well for 16 h. Cells were preincubated with various concentrations of chloroquine for 30 min, followed by stimulation with 10 μ g/ml CpG (NBP2-26232) for 24 h. SEAP was analyzed using SEAPorter™ Assay Kit (NBP2-25285). Dose-responsive percent inhibition of each sample well was calculated to yield the chloroquine IC_{50} value.

Reference:

- Macfarlane, D. E. et al. (1998). Antagonism of immunostimulatory CpG-oligodeoxynucleotides by quinacrine, chloroquine, and structurally related compounds. *J. Immunol.* 160, 1122-1131.
- Rutz, M. et al. (2004). Toll-like receptor 9 binds single-stranded CpG-DNA in a sequence- and pH-dependent manner. *Eur. J. Immunol.* 34, 2541-2550.
- Marshak-Rothstein, A. (2006). Toll-like receptors in systemic autoimmune disease. *Nat. Rev. Immunol.* 6, 823-835.