LPS-EB is a preparation of a smooth (s)-form of lipopolysaccharide (LPS) purified from the Gram-negative *E. coli* 0111:B4, a pathogenic serotype of *E. coli* known to cause significant gastric disease. LPS-EB Vaccigrade™ is extracted by successive enzymatic hydrolysis steps and purified by the previously described phenol-TEA-DOC extraction protocol. This process removes contaminating lipoproteins. Therefore, LPS-EB Vaccigrade™ only activates TLR4, as verified using InvivoGen’s HEK-Blue™ hTLR2 and HEK-Blue™ hTLR4 cells (Figure 1).

Figure 1. **LPS-EB Vaccigrade™** is a potent activator of human (h)TLR4. The cells were incubated with increasing concentrations of LPS-EB Vaccigrade™. After overnight incubation in HEK-Blue™ detection medium, a SEAP detection growth medium, the response of hTLR2 and hTLR4 was assessed by determining the presence of SEAP in the supernatant. Data are expressed as optical density at 630 nm (±SEM).

*Note:* 1 mg/ml corresponds to $1 \times 10^6$ EU/ml.