



Florfenicol ELISA Kit

11/19

(Catalog # E4780-100; 96 assays, Storage at 4°C)

I. Introduction:

Florfenicol is a broad-spectrum, primarily bacteriostatic, antibiotic with a range of activity similar to that of chloramphenicol, including many gram-negative and gram-positive organisms. It has a same mechanism of action as chloramphenicol i.e. inhibition of protein synthesis. Signs of Florfenicol toxicity are varied and include diarrhea and hyperbilirubinemia in horses, diarrhea and decreased feed consumption and rumen activity in cattle. Overdose or prolonged florfenicol administration can cause fatal bone marrow suppression. Florfenicol ELISA Kit is based on Competitive ELISA principle. The micro-plate provided in this kit has been pre-coated with Florfenicol. During the reaction, Florfenicol in the samples or standard competes with Florfenicol coated on the plate for binding to the anti-Florfenicol antibody. Then Horseradish Peroxidase (HRP) conjugate is added to each micro plate well, and TMB substrate is for color development. There is a negative correlation between the OD value of samples and the concentration of Florfenicol. The concentration of Florfenicol in the samples can be calculated by comparing the OD of the samples to the standard curve.

II. Applications:

In vitro, quantitative determination of Florfenicol

Sensitivity: 0.15 ppb (ng/mL)

Detection Range: Tissue, Liver, Honey, Milk - 0.075ppb; Feed, Milk powder - 0.15 ppb

Sample recovery rate: Tissue, Liver - 85%±20%; Honey- 85%±25%; Feed, Milk - 75%±25%.

Cross-reactivity: Florfenicol -100%; Thiamphenicol - 0.1%

III. Sample Type:

Tissue, Honey

IV. Kit Contents:

Components	E4780-100	Part Number
Micro ELISA Plate	96 wells	E4780-100-1
Standard	6 X 1 ml	E4780-100-2
HRP Conjugate	5.5 ml	E4780-100-3
Antibody Working Solution	5.5 ml	E4780-100-4
Substrate Reagent A	6 ml	E4780-100-5
Substrate Reagent B	6 ml	E4780-100-6
Stop Solution	6 ml	E4780-100-7
Wash Buffer (20X)	40 ml	E4780-100-8
Reconstitution Buffer (2X)	50 ml	E4780-100-9
Plate Sealer	3	E4780-100-10

V. User Supplied Reagents and Equipment:

- Microplate reader capable of measuring absorbance at 450 nm
- Ethyl acetate, N-hexane, Acetonitrile, Na₂Fe(CN)₅(NO)•2H₂O, ZnSO₄•7H₂O
- Clean Eppendorf tubes for preparing standards or sample dilutions

VI. Storage and Handling:

Store at 4°C.

VII. Reagent and Sample Preparation:

Bring all reagents to room temperature before use. Before using the kit, spin tubes and bring down all components to the bottom of tubes.

- **Wash Buffer (20X):** Dilute 20X Concentrated Wash Buffer to 1X with deionized water.
- **Reconstitution Buffer (2X):** Dilute 2X Reconstitution Buffer with deionized water. Mix 2x Reconstitution Buffer (V): Deionized water (V) =1:1). The Reconstitution buffer can be store at 4°C for a month.
- **0.36M Na₂Fe (CN)₅(NO)•2H₂O Solution:** Dissolve 10.7 g of Na₂Fe(CN)₅(NO)•2H₂O to 100 mL with deionized water.
- **1.04M ZnSO₄•7H₂O Solution:** Dissolve 29.8 g of ZnSO₄•7H₂O to 100 mL with deionized water.
- **Acetonitrile -Water Solution:** Acetonitrile (V): Deionized water (V) =84:16
- **Standard:**

Standard	S1	S2	S3	S4	S5	S6
Concentration (ppb)	0	0.15	0.45	1.35	4.05	12.15

