# CHLORAMPHENICOL

## IDENTITY

**Chemical name:** 2,2-dichloro-N-[2-hydroxy-1-(hydroxymethyl)-2-(4-nitrophenyl) ethyl] acetamide

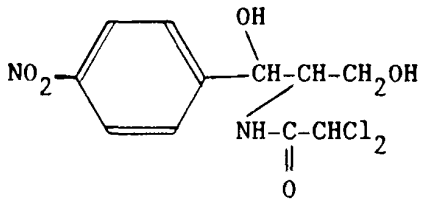
D-threo-N-(1,1'-dihydroxy-1-p-nitrophenylisopropyl) dichloroacetamide

D(-)-threo-2-dichloroacetamido-1-p-nitrophenyl-1,3-propanediol

D-threo-N-dichloroacetyl-1-p-nitrophenyl-2-amino-1,3-propanediol

**Synonyms:** Sold under at least 66 trade names

**Structural formula:**




**Molecular formula:** $C_{11}H_{12}Cl_2N_2O_5$

**Molecular weight:** 323.14

## OTHER INFORMATION ON IDENTITY AND PROPERTIES

**Pure active ingredient:**

**Appearance:** Needles or elongated plates

**Melting Point:** 150.5-151.5°C

**Optical Rotation:** $[\alpha]_D^{27}$ = + 18.6°C (c = 4.86 in ethanol)

**$UV_{max}$:** 278 nm

(Windholz, 1983)

**Technical Active Ingredients:**

Chloramphenicol is obtained by isolation from the soil bacteria Streptomyces venezuela. Alternatively, chloramphenicol may be prepared by several synthetic methods. (Windholz, 1983)

## RESIDUES IN FOOD AND THEIR EVALUATION

### CONDITIONS OF USE:

**General:**

Chloramphenicol is a broad spectrum antibiotic particularly effective against gram negative bacteria including salmonella, pasteurella and coliforms (Milhaud, 1985). It is considered a potent antibiotic for treating pneumonic and enteric conditions (Mercer, 1980). Chloramphenicol has been recommended for prevention of secondary infections associated with chronic respiratory disease in poultry (Sisodia and Dunlop, 1972a, 1972b).

In some countries, the use of chloramphenicol in food-producing animals is prohibited; in others, it can only be obtained by prescription from licensed veterinarians.

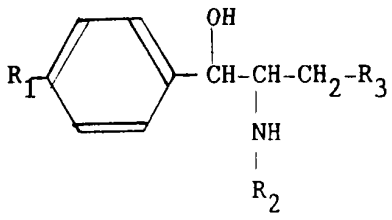
**Dosages:**

Chloramphenicol can be prepared as an injectable suspension in water, ethanol or propylene glycol in dose ranges of 22-66 mg/ml. Other routes of administration include oral (tablets and in drinking water), topical ointments, and ophthalmic solutions. Dosage formulations also are available for chloramphenicol succinate and chloramphenicol palmitate.

### RADIOLABELED METABOLISM STUDIES:

Several metabolites of chloramphenicol have been identified in rat urine following intramuscular injection. Metabolites identified include free chloramphenicol and its glucuronide, the oxamic acid derivative and its corresponding alcohol, the reduced arylamine and the acetylarylamine. Based on radioactivity recoveries, the major metabolites appear to be chloramphenicol base (27%) and the acetylarylamine (19%). Other metabolites were recovered at 8 to 15 percent except the arylamine (4%). These metabolites are shown in Table I (Bories, et al., 1983a). In vitro studies with pig liver showed activity similar to that of the rat. The same experiments with sheep and cattle liver preparations showed these to have much lower glucuronyl transferase activities (25% and 19%, respectively) (Smith, et al., 1984). No radiolabeled dosing studies were found for cattle and dairy cows, pigs, sheep, or poultry.

Table I. Metabolites of Chloramphenicol in the Rat



| | Compound | $R_1$ | $R_2$ | $R_3$ |
|---|---|---|---|---|
| 1. | chloramphenicol | $NO_2$ | $COCHCl_2$ | OH |
| 2. | glucuronide | $NO_2$ | $COCHCl_2$ | $C_6H_9O_7$ |
| 3. | oxamic acid | $NO_2$ | COCHOOH | OH |
| 4. | alcohol | $NO_2$ | $COCH_2OH$ | OH |
| 5. | base | $NO_2$ | H | OH |
| 6. | acetylarylamine | $NHCOCH_3$ | $COCHCl_2$ | OH |
| 7. | arylamine | $NH_2$ | $COCHCl_2$ | OH |

**Residue Studies:**

A variety of depletion studies have been conducted. However, it should be noted that except where a glucuronidase hydrolysis procedure was included in the method of analysis, the methods detected only the parent chloramphenicol compound.

Eggs from laying hens treated orally with a 10% chloramphenicol solution (50 mg/kg every 12 hours for 3 days) gave 8000 µg/kg at day 5, 15 µg/kg at day 10 and 3 µg/kg at day 15 in egg whites. Concentrations in egg yolks were 1500 µg/kg at day 1, 8 µg/kg at day 5 and less than 1 µg/kg at day 7 (Boisseau, 1987).

Chloramphenicol residues in calves have been reported using a gas chromatographic method (Epstein et al., 1986). This study used chloramphenicol in propylene glycol at 66 mg/kg body weight (b.w.) by intravenous (IV) injection and at 33 mg/kg b.w. and 66 mg/kg b.w. by intramuscular (IM) injection. All animals were dosed twice within a 24 hour interval. Calves were sacrificed at designated times from 2 to 72 hours post-treatment. Results have been tabulated in Table II. The half-life for the IM doses was approximately 7 hours. This is approximately double the 3 hour half-life of the IV dosed calves.

Table II. Tissue Residues in Calves

| Group | Withdrawal Times (Hrs) | Chloramphenicol Concentration (mg/kg) Injection Site A | Injection Site B | Shoulder A | Shoulder B | Gluteal A | Gluteal B |
|---|---|---|---|---|---|---|---|
| 1 (33 mg/kg) i.m. | 2 | 918 | 911 | 3.60 | 6.72 | 2.67 | 1.70 |
| | 6 | 296 | 1030 | 5.22 | 7.03 | 3.26 | 11.40 |
| | 24 | 408 | 168 | 0.203 | 3.90 | 0.382 | 4.71 |
| | 48 | 13.2 | 10.2 | 0.185 | 0.749 | 0.162 | 0.843 |
| | 72 | 19.4 | 1.77 | 0.360 | 1.43 | 7.68 | 0.807 |
| 2 (66 mg/kg) i.m. | 2 | 3390 | 2250 | 3.36 | 9.86 | 7.18 | 8.60 |
| | 4 | 756 | 1490 | 8.85 | 13.0 | 6.64 | 7.54 |
| | 24 | 444 | 333 | 7.53 | 7.00 | 8.67 | 7.76 |
| | 48 | 270 | 259 | 0.205 | 0.204 | 0.375 | 0.76 |
| | 72 | 1.77 | 23.9 | 0.218 | 1.09 | 0.449 | 0.823 |
| 3 (66 mg/kg) i.v. | 2 | 75.9 | NA | 68.2 | NA | 74.4 | NA |
| | 4 | 33.6 | NA | 33.6 | NA | 32.6 | NA |
| | 24 | 0.29 | NA | 0.174 | NA | 0.371 | NA |
| | 48 | 0.122 | NA | 0.371 | NA | 0.079 | NA |

A and B are designations for each calf killed and evaluated at each hour after the last chloramphenicol dose; different calves were involved at each interval. NA = Not analyzed.

## METHODS OF RESIDUE ANALYSIS

Recent reviews of analytical methods for chloramphenicol residues include those published by Arnold and Somogyi (1982) and Milhaud (1985). Allen (1985) has published a review evaluating gas, liquid and thin layer chromatographic procedures. Results from many laboratories have been reported using these procedures, but only a very limited number have used multi-laboratory collaborated methods with established performance characteristics. Results from most studies may, therefore, be inconclusive but they do provide general information on chloramphenicol residues.

A modification of the U.S. Department of Agriculture method (USDA, 1986) for urine and muscle includes the use of a glucuronidase enzyme digestion step to convert the glucuronide metabolite to the free chloramphenicol. The significance of this modification is that not only is free chloramphenicol detected, but also at least one metabolite is hydrolyzed to the free chloramphenicol so that it can be quantitated as well. This is important as the presence of this enzyme in the human digestive system would readily hydrolyze this metabolite to the parent compound.

Chemical methodology provides for assays that are able to detect chloramphenicol to the 10 µg/kg level and below. Wal et al., (1980) have reported on an HPLC procedure for milk with quantitation at 10 µg/kg. Bories et al., (1983b) published a paper on the liquid chromatographic determination and mass spectrometric confirmation of chloramphenicol residues in animal tissues down to 10 µg/kg.

## APPRAISAL

The use of chloramphenicol in food-producing animals results in residues of the drug in the meat, milk and eggs. Chemical methods are available for quantitating the parent drug at levels of at least 10 µg/kg.

Improvements in analytical technology promise to provide methods with improved sensitivity without sacrificing other performance characteristics. Because of the potential for the significant relative contribution of the glucuronide metabolite in various assays, future methods development and the application of current methods should consider the inclusion of an enzyme hydrolysis step.

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