Date of Approval: June 9, 2023

CORRECTED FREEDOM OF INFORMATION SUMMARY

SUPPLEMENTAL ABBREVIATED NEW ANIMAL DRUG APPLICATION

ANADA 200-117

Oxytetracycline Injection

Injectable solution

Beef cattle, non-lactating dairy cattle, and swine

Provides information to address the human food safety and user safety of *N*-methyl-2-pyrrolidone (NMP) in the formulation of Oxytetracycline Injection, and provides for alignment with Guidance for Industry (GFI) #263, "Recommendations for Sponsors of Medically Important Antimicrobial Drugs Approved for Use in Animals to Voluntarily Bring Under Veterinary Oversight All Products That Continue to be Available Over-the-Counter."

Sponsored by:

Bimeda Animal Health Ltd.

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I. GENERAL INFORMATION

A. File Number

ANADA 200-117

B. Sponsor

Bimeda Animal Health Ltd.
1B The Herbert Building, The Park
Carrickmines, Dublin 18, Ireland

Drug Labeler Code: 061133

U.S. Agent Name and Address:

Deb Ann Voss
Bimeda Inc.
291 Forest Prairie Road
Le Sueur, MN 56058

C. Proprietary Name

Oxytetracycline Injection

D. Drug Product Established Name

oxytetracycline injection

E. Pharmacological Category

Antimicrobial

F. Dosage Form

Injectable solution

G. Amount of Active Ingredient

200 mg/mL of oxytetracycline base as oxytetracycline dihydrate

H. How Supplied

250 and 500 mL vials

I. Dispensing Status

Prescription (Rx)

J. Dosage Regimen

There were no changes to the dosage regimen as a result of this supplemental approval.

K. Route of Administration

Intramuscular injection (cattle and swine) and intravenous injection (cattle)

L. Species/Class

Beef cattle, non-lactating dairy cattle, and swine

M. Indications

Oxytetracycline Injection is intended for use in the treatment of the following diseases in beef cattle, non-lactating dairy cattle, and swine when due to oxytetracycline susceptible organisms:

Cattle:
In cattle, Oxytetracycline Injection is indicated in the treatment of pneumonia and shipping fever complex associated with *Pasteurella* spp. and *Hemophilus* spp.; infectious bovine keratoconjunctivitis (pinkeye) caused by *Moraxella bovis*; foot rot and diphtheria caused by *Fusobacterium necrophorum*; bacterial enteritis (scours) caused by *Escherichia coli*; wooden tongue caused by *Actinobacillus lignieresii*; leptospirosis caused by *Leptospira pomona*; and wound infections and acute metritis caused by strains of staphylococci and streptococci organisms sensitive to oxytetracycline.

Swine:
In swine, Oxytetracycline Injection is indicated in the treatment of bacterial enteritis (scours, colibacillosis) caused by *Escherichia coli*; pneumonia caused by *Pasteurella multocida*; and leptospirosis caused by *Leptospira pomona*. In sows, Oxytetracycline Injection is indicated as an aid in the control of infectious enteritis (baby pig scours, colibacillosis) in suckling pigs caused by *Escherichia coli*.

N. Reference Listed New Animal Drug (RLNAD)

Liquamycin® LA-200®; oxytetracycline injection; NADA 113-232; Zoetis Inc.

O. Effect of Supplement

This supplement provides information to address the human food safety and user safety of *N*-methyl-2-pyrrolidone (NMP) in the formulation of Oxytetracycline Injection, and provides for alignment with Guidance for Industry (GFI) #263, "Recommendations for Sponsors of Medically Important Antimicrobial Drugs Approved for Use in Animals to Voluntarily Bring Under Veterinary Oversight All Products That Continue to be Available Over-the-Counter."

II. BIOEQUIVALENCE

CVM did not require additional bioequivalence information for this supplemental approval. The Freedom of Information (FOI) Summary for the original approval of ANADA 200-117, dated April 13, 1995, contains a summary of data that demonstrate bioequivalence of Oxytetracycline Injection for cattle and swine.

III. HUMAN FOOD SAFETY

The following are assigned to this product for beef cattle, non-lactating dairy cattle, and swine:

A. Toxicology

Oxytetracycline
The acceptable daily intake (ADI) for total tetracycline residues (chlortetracycline, oxytetracycline, and tetracycline) is 25 micrograms per kilogram of body weight per day (µg/kg bw/day) as codified under 21 CFR 556.500.

No additional toxicology studies were required for oxytetracycline for this supplemental approval.

N-methyl-2-pyrrolidone (NMP)

1. Summary of Toxicology Studies

   The following studies, together with the studies in the original approval of ANADA 200-117, dated April 13, 1995, were considered in establishing the mode of action (how NMP is thought to cause cancer in mice) and threshold (the amount of NMP that will not cause cancer in mice) for liver carcinogenesis of NMP, an excipient used in Oxytetracycline Injection.

   a. Genotoxicity Studies

      (1) Bacterial Reverse Mutation Assay (Ames Test)

      Title: Mutagenicity Test on GMP M-Pyrol (93519-90) in the *Salmonella*/Mammalian Microsome Reverse Mutation Assay (Ames Test) with a Confirmatory Assay. (Study No. 12596-0-401R)

      Study Dates: January 14, 1991 to January 9, 1992

      Study Location: Kensington, Maryland

      Study Design: This assay evaluated the test article and/or its metabolites for their ability to induce reverse mutations at the histidine locus in the genome of specific *Salmonella typhimurium* tester strains both in the presence and absence of an exogenous metabolic activation system of mammalian microsomal enzymes derived from Aroclor-induced rat liver (S9). The tester strains used were the *S. typhimurium* histidine auxotrophs TA98, TA100, TA1535, TA1537, and TA1538. The doses tested in the mutagenicity assay were selected based upon the results of a dose range finding study using tester strain TA100 and ten dose levels of the test article ranging from 6.67 to 5000 µg per plate, one plate per dose, both in the presence and absence of S9. The confirmatory assay was conducted using three plates per dose level both in the presence and absence of S9. Six doses of the test article, ranging from 100 to 500 µg per plate, were tested.

Results and Conclusion: In either the initial or confirmatory mutagenicity assay, no positive increases in the number of histidine revertants per plate were observed with any of the tester strains in the presence or absence of S9. NMP was not mutagenic under the conditions of this assay.

(2) *In Vivo* Micronucleus Test

Title: Cytogenetic Study *In Vivo* of N-Methylpyrrolidinone in Mice, Micronucleus Test, Single Oral Administration. (Study No. 26M0369/884156)

Study Dates: August 24, 1988 to March 7, 1989

Study Location: Ludwigshafen, Germany

Study Design: The *in vivo* micronucleus test in mice was to test the potential of NMP to cause genetic damages in treated animals. The study was conducted according to the Organization for Economic Co-operation and Development (OECD) Guideline for the Testing of Chemicals No. 474 (1983). NMP (purity >99.8%) was administered as a single dose to adult male and female NMRI mice through gavage in a volume of 10 mL/kg bw at the dose levels of 950, 1900, 3800 mg/kg bw (low, mid, and high dose, respectively). Water was used as the negative control as well as dosing vehicle for NMP and the positive controls. Cyclophosphamide (40 mg/kg bw) was used as the positive control for clastogenic effects, and vincristine (0.15 mg/kg bw) was used as the positive control for spindle poison effects. The group size was 5 animals/sex/treatment/time-point for the vehicle control and NMP-treatment groups; a total of five mice (males and females) were used for each of the positive control groups. The mice in the vehicle control, positive control, and low and mid dose groups were sacrificed at 24 hours post-dose for sampling; the sampling times for the high dose group were 16-, 24-, and 48-hours post-dose. Bone marrow samples were harvested from both femurs and smeared on glass slides for staining and evaluation according to standard procedures. A total of 1000 polychromatic erythrocytes (PCE) per animal were scored for incidence of micronuclei (MN). The ratio of PCE to normochromatic erythrocytes (NCE) was determined for each animal based on the counting of 1000 erythrocytes. In addition, the number of small and large micronuclei were counted.

Results and Conclusion: Mice treated with the dosing vehicle and the positive controls did not show any clinical signs of toxicity, whereas the NMP-treated animals, at all doses, showed irregular respiration and general stress following administration. While both positive controls showed significantly higher MN counts (approximately 9- and 50-fold increases compared to the vehicle control), there was no difference between the NMP-treated and the vehicle control animals in the number of MN. The NCE/PCE ratio was higher at the 24- and 48-hour time points for the high dose group than that of the vehicle control, indicating possible bone marrow toxicity.

The results of this study indicated that under the experimental conditions, treatment with NMP *in vivo* did not increase the formation of micronuclei in the bone marrow erythrocytes of the treated mice.

b. *In Vivo* Mechanistic Study – PPARα and CAR activation

Title: A 1-week Study of N-Methyl-2-Pyrrolidone (NMP) by Oral Gavage Administration in Mice. (Study No. 20032029)

Study Dates: October 10, 2012 to September 24, 2013

Study Location: Spencerville, Ohio

Study Design: B6C3F1 mice (5 animals/sex/dose) were orally administered by gavage the test article, NMP, at 0, 1000, and 3000 mg/kg bw/day for 7 consecutive days to evaluate the mode-of-action for NMP-induced hepatic carcinogenesis in this Good Laboratory Practices (GLP) study. Trichloroacetic acid (TCA), a positive control Peroxisome Proliferator Activated Receptor Alpha (PPARα) agonist group (5 animals/sex), was administered in drinking water at 300 mg/kg bw/day. General observations and clinical signs were performed. Body weight and water consumption were recorded. At termination of the study, liver tissue samples were collected from all animals for biochemical and molecular analyses, necropsy examinations were performed, and organ weights for brain and liver were obtained. Histopathology evaluations were conducted on livers from all animals.

Results and Conclusion: All mice survived until termination of the study. Decreased activity, few feces, urine staining, dehydration, and distended abdomen were noted at the high dose of NMP (3000 mg/kg bw/day). An absolute mean body weight decrease (~9%) was noted at both 1000 and 3000 mg/kg bw/day for females. No body weight changes in males occurred during the study. There were treatment-related increases in liver weight, both absolute and relative to brain or body weight, in females at 3000 mg/kg bw/day. Centrilobular hypertrophy of the hepatocytes and increased mitotic figures were observed in all animals in the 3000 mg/kg bw/day group. Males showed an increase in the number of hepatocytes undergoing mitosis in the 1000 mg/kg bw/day dose group. NMP treatment (3000 mg/kg bw/day) resulted in a 10.9-fold induction of RNA expression of cytochrome P450 2b10 (indicator of Constitutive Androstane Receptor (CAR) mediated response) in males and 4.3-fold in females. RNA expression of cytochrome P450 4a10 (indicator of PPARα mediated response) was induced 2.9-fold in males and unchanged in females at 3000 mg/kg bw/day. The positive control, TCA, a known hepatocarcinogen, produced expected results of PPARα activation and little or no change in indicators of CAR activation. These results provide evidence that activation of PPARα and CAR may lead to induced cellular proliferation, liver hypertrophy with Cyp2b induction, increased mitotic figures and liver weights, which are some of the key events in the progression of NMP-induced hepatocarcinogenesis in mice.

c. Subchronic Oral Toxicity Study in Rodents

Title: 90-Day Subchronic Toxicity Study in Rats and Mice Fed N-Methylpyrrolidone (NMP) Including Neurotoxicity Evaluation in Rats. (Published article in Drug and Chemical Toxicology, 22(3): 455-480 (1999))

Study Location: Ludwigshafen, Germany

Study Design: NMP was administered in the diet for 90 days to groups of 10 male and 10 female B6C3F1 mice at a diet concentration of 0, 1000, 2500, or 7500 ppm (the concentration in ppm was estimated to be equivalent to 0, 167, 417, or 1250 mg/kg bw/day, to account for a lack of accurate food consumption data due to spillage from clumping which impeded proper feeder flow). Body weight, body weight gain, ophthalmologic examination, clinical chemistry, hematology, organ weights, histopathological examination, and clinical observations were obtained. A satellite 28-day study with 10 male and 10 female mice was performed, as well as, a 90-day study in Crl:CD®BR rats, but are not summarized here because the data were not used for the carcinogenic assessment of NMP.

Results and Conclusion: No compound-related effects on mortality, hematologic parameters, body weight, or food consumption were reported. Changes in urine color, but not kidney function were observed at the doses of 417 and 1250 mg/kg bw/day. Changes in cholesterol, triglycerides, calcium, and alkaline phosphatase at 28 days, but not following 90 days of NMP treatment were reported. Liver weights were elevated in males at 417 mg/kg bw/day and in both males and females at 1250 mg/kg bw/day for 90 days. Hepatocellular hypertrophy was noted in both males and females at 417 and 1250 mg/kg bw/day. The no-observed-effect level (NOEL)/no-observed-adverse-effect level (NOAEL) for this study was 167 mg/kg bw/day based on the increased liver weight and increased incidences of centrilobular hepatocellular hypertrophy observed at 417 mg/kg bw/day.

d. Oral Carcinogenicity Study in Mice

Title: Chronic Toxicity and Oncogenicity of *N*-methylpyrrolidone (NMP) in Rats and Mice by Dietary Administration. (Published article in Drug and Chemical Toxicology, 24(4): 315-338 (2001))

Study Location: Ludwigshafen, Germany

Study Design: NMP was administered in the diet for at least 18 months (78 to 80 weeks) to groups of 50 male and 50 female B6C3F1/CrlBR mice at a diet concentration of 0, 600, 1200, or 7200 ppm (the concentration in ppm was estimated to be equivalent to 0, 100, 200, or 1200 mg/kg bw/day, to account for the lack of accurate food consumption data from spillage). Body weight, body weight gain, clinical chemistry, hematology, organ weights, histopathological examination, and clinical observations were obtained. A 2-year study in Crl:CD® (SD)BR rats was also described, but is not summarized here because the data were not used for the carcinogenic assessment for NMP.

Results and Conclusion: Administration of the test article resulted in an increase in the incidence of liver tumors and foci of cellular alteration in the liver of both male and female mice receiving the highest dose. Centrilobular hypertrophy of the hepatocytes was noted in most males in this group and correlated with the significant increase in liver weight changes. At 1200 ppm, relative liver weights were increased in the males, and three males had centrilobular hypertrophy. The effects in the liver were considered a consequence of enzyme induction.

In conclusion, under the conditions of this study, tumorigenic effects were associated with the administration of the test article, NMP. The incidences of both benign and malignant liver tumors and preneoplastic liver lesions (foci of cellular alteration) were increased in male and female mice receiving 7200 ppm compared to control mice. A NOEL/NOAEL of 600 ppm (100 mg/kg bw/day) is established for this study based on centrilobular hypertrophy of hepatocytes and renal lipid vacuole formation observed in mid- and high-dose male mice.

2. Summary of Carcinogenic Mode of Action

According to 21 CFR 500.84(c), FDA evaluated an alternate procedure proposed by the sponsor as provided in 21 CFR 500.90 to determine the concentration of a residue of carcinogenic concern in the total human diet that represents no significant increase in the risk of cancer to the human consumer. The sponsor has petitioned a waiver of requirements according to 21 CFR 500.90 and has explained the reasons why this alternative procedure provides the basis for concluding that approval of the compound satisfies the requirements of the anticancer provisions of the Federal Food, Drug, and Cosmetic Act. Based on the non-genotoxic mode of action summarized below, the alternative approach to the regulation for carcinogenic residues of NMP in the human diet is scientifically acceptable.

NMP was not genotoxic in a battery of genotoxicity studies. However, NMP administration resulted in increased liver weight and hepatocellular hypertrophy and induced liver tumors in B6C3F1/CrlBR mice in an 18-month dietary oncogenicity study. The pleiotropic effects of increased liver weight and hepatocellular hypertrophy are known hallmarks of PPARα and CAR activation. The weight of evidence from the studies listed above supports both CAR and PPARα mediated responses as a non-genotoxic mode of action for NMP-induced carcinogenesis in the mouse. Based on the results of the mechanistic work, CAR activation appears to be the predominant mode of action whereas activation of PPARα is observed to a lesser extent. The strongest evidence comes from the one-week mouse study, in which the elevated liver weights and centrilobular hepatocyte hypertrophy is consistent with the hepatic enzyme induction observed in the subchronic study in mice described above (see Section III.A.1). Also, the 1-week mouse study demonstrated that NMP may induce activation of PPARα and CAR transcriptional responses by measuring changes in receptor-regulated gene expression after dosing. These results clearly show that NMP induces cytochrome P450 2b10 (Cyp2b10) in male and female mice, a finding consistent with a CAR nuclear receptor-mediated response. Also, the minimal cytochrome P450 4a10 increase observed in male mice is compatible with a weaker PPARα response. The

observed hepatic enzyme induction is consistent with what is commonly reported in activated PPARα and CAR pathways.

Thus, progression of key events for the carcinogenic mode of action associated with the mouse-CAR toxicological-signal would be: mouse-CAR activation, altered gene expression, cell proliferation, clonal expansion leading to altered foci, mouse liver tumor. Associated events would include hypertrophy, Cyp2b10 induction and altered apoptosis. The induction of mouse liver tumors by NMP is consistent with the list of key events in the scientific literature on the mode of action, including mouse-PPARα activation, cell proliferation and altered apoptosis, preneoplastic foci, clonal expansion, mouse liver tumors. Associated events would include expression of peroxisomal genes, an increase in peroxisome numbers, and an increase in peroxisome size.

3. Determination of the Residue of Carcinogenic Concern in the Total Human Diet (S_o) for NMP

   The S_o is defined as the concentration of a residue of carcinogenic concern in the total human diet that represents no significant increase in the risk of cancer to the human consumer.

   Based on the available toxicology studies, the NOEL/NOAEL of 167 mg/kg bw/day from the 90-day oral toxicity study in mice [Drug and Chemical Toxicology, 22: 455-480 (1999)] was selected to be the most appropriate point of departure for determining the S_o for chronic human exposure to the NMP residue of carcinogenic concern. The NOEL/NOAEL of 167 mg/kg bw/day is a dose of NMP that does not cause observable adverse effects when orally administered to mice for 90 days.

   The S_o for the residue of NMP is calculated using the following formula based on the NOEL/NOAEL of 167 mg/kg bw/day from the subchronic oral toxicity study in mice as the point of the departure, a safety factor of 100, a total diet in humans of 1.5 kg/day, and assuming a 60-kg average human body weight. The safety factor of 100 was applied to account for a 10-fold factor for study duration and a 10-fold factor for human-to-human variability and takes into account that the rodent is either uniquely sensitive to PPAR- and CAR-mediated carcinogenicity, or at least considerably more sensitive than other mammalian species.

$$S_o = \frac{\text{Point of Departure/Safety Factor} \times \text{Human Body Weight}}{\text{Total Food in the Human Diet}}$$

$$= \frac{167 \text{ mg/kg bw/day}/100 \times 60 \text{ kg bw}}{1.5 \text{ kg/day}}$$

$$= 66.8 \text{ mg/kg food in the total diet (66.8 ppm)}$$

The S_o for the NMP residue of carcinogenic concern in the total human diet is 66.8 ppm.

B. S_m for the NMP Residue of Carcinogenic Concern in Edible Tissues

The S_m is the concentration of a residue of carcinogenic concern in a specific edible tissue corresponding to no significant increase in the risk of cancer to the human consumer; FDA will assume that this S_m will correspond to the concentration of residue in a specific edible tissue that corresponds to a maximum lifetime risk of cancer in the test animals of 1 in 1 million (21 CFR 500.82(b) and 500.84(c)(1)). Because not all of the total human diet (1.5 kg/day, or 1500 g/day) is derived from food-producing animals, a correction for food intake is made in determining the S_m. The S_m is calculated based on the S_o (in this case, 66.8 ppm), assuming that up to 500 g/day of the total diet is due to the consumption of meat. Of this 500 g total daily meat consumption, 300 g is assumed to be comprised of muscle, 100 g comprised of liver, 50 g comprised of kidney, and 50 g comprised of fat. Skin with fat in natural proportions is the edible tissue for swine, however, in this document; we refer to fat for both cattle and swine as the edible tissue for simplicity. Thus, $^1/_5$ of the total diet (300 g muscle/1500 g total diet), would be comprised of muscle, $^1/_{15}$ comprised of liver, $^1/_{30}$ comprised of kidney and $^1/_{30}$ comprised of fat. For example, the concentration of the NMP residue of carcinogenic concern in muscle, S_m muscle, would be calculated as 66.8 ppm ÷ $^1/_5$, which equals 334 ppm.

Calculation of the S_m for the individual edible tissues (as summarized in Table III.1) is as follows:

S_m muscle $= 66.8 \text{ ppm} \div \left(\frac{1}{5}\right) = 334 \text{ ppm}$

S_m liver $= 66.8 \text{ ppm} \div \left(\frac{1}{15}\right) = 1002 \text{ ppm}$

S_m kidney $= 66.8 \text{ ppm} \div \left(\frac{1}{30}\right) = 2004 \text{ ppm}$

S_m fat $= 66.8 \text{ ppm} \div \left(\frac{1}{30}\right) = 2004 \text{ ppm}$

Table III.1. Summary Table of the NMP Residue of Carcinogenic Concern in Edible Tissues of Cattle and Swine

Edible Tissue	Amount Consumed Per Day	Fraction of Total Diet	S_m Concentration
Muscle	300 g	1/5	334 ppm
Liver	100 g	1/15	1002 ppm
Kidney	50 g	1/30	2004 ppm
Fat	50 g	1/30	2004 ppm

C. Metabolism and Total Residue Studies for NMP in Cattle and Swine

Because the depletion characteristics of the excipient, NMP, used in the generic product were not known, abbreviated total residue studies were conducted to determine whether the generic product would be regulated on the basis of NMP, or the active ingredient, oxytetracycline.

1. Total Residue Study in Cattle

   Title: Abbreviated Total Residue and Metabolism Study for ^{14}C-N-Methylpyrrolidone Administered to Beef Cattle. (Study No. ADC Projects 1102 and 1102A)

   Study Dates: March 2, 1989, to January 31, 1992

   Study Location: Colorado Springs and Fort Collins, Colorado

   Study Design and Results: Six beef-type, steers and heifers 7 to 8 months of age were used. A single dose of [^{14}C]-NMP was administered intramuscularly in three portions. The tissues were analyzed for total radioactive residues of [^{14}C]-NMP. The data indicate that liver is the target tissue for NMP in cattle although total residues of NMP in liver and kidney were very similar. At one-day withdrawal, the NMP total radioactive residue concentrations in all tissues were substantially below their respective S_m values.

   Table III.2. Mean (± standard deviation) total residues of NMP equivalents (ppm) in tissues of cattle administered a single intramuscular injection of 41.32 mg [^{14}C]-NMP/kg bw in three portions.

Withdrawal Period (days)	Muscle	Kidney	Liver	Fat	Injection Site
1	19.93±0.84	28.04±4.82	23.43±1.12	2.42±0.66	20.82±0.42
4	1.67±0.34	3.42±0.64	4.86±1.09	0.25±0.01	1.65±0.30
21	0.35±0.01	0.42±0.01	0.78±0.01	0.20±0.00	0.31±0.04

2. Total Residue Study in Swine

   Title: Abbreviated Total Residue and Metabolism Study for ^{14}C-N-Methylpyrrolidone Administered to Swine. (Study No. ADC Project 1254)

   Study Dates: April 25, 1991, to August 10, 1992

   Study Location: Colorado Springs and Fort Collins, Colorado

   Study Design and Results: Seven Hampshire cross, barrows and gilts weighing approximately 50 pounds were used. A single dose of [^{14}C]-NMP was administered intramuscularly. The tissues were analyzed for total radioactive residues of [^{14}C]-NMP. The data indicate that liver is the target tissue for NMP in swine although total residues of NMP in liver and kidney were very similar. At one-day withdrawal, the NMP total radioactive residue concentrations in all tissues were substantially below their respective S_m values.

Table III.3. Mean (± standard deviation) total residues of NMP equivalents (ppm) in tissues of swine administered a single intramuscular injection of 41.32 mg [^{14}C]-NMP/kg bw.

Withdrawal Period (days)	Muscle	Kidney	Liver	Fat	Injection Site
1	18.30±1.27	26.55±0.21	21.75±3.32	5.20±0.75	15.15±4.17
4	0.39	0.72	0.23	0.29	0.46
21	0.07±0.03	0.05±0.01	0.14±0.01	0.07±0.02	0.07±0.02

Day 3 data are from a dead animal and are not included in the table.

The results of total residue studies in cattle and swine demonstrated that residues of NMP would deplete to the S_m before 28 days withdrawal. Because this is less than the 28-day withdrawal for oxytetracycline codified under 21 CFR 522.1660a, the generic product is regulated based on the depletion of the active ingredient, oxytetracycline.

3. Comparative Metabolism Study

Title: Comparative metabolism study of ^{14}C-N-methyl-2-pyrrolidone Administered Orally to Rats. (Study No. ADC Projects 1102 and 1102A)

Study Dates: July 19, 1991, to January 31, 1992

Study Location: Colorado Springs, Colorado

Study Design and Results: Eleven male and eleven female Sprague-Dawley rats weighing approximately 250 g (males) and 225 g (females) were used. [^{14}C]-NMP was administered by oral gavage at a nominal rate of 50 mg/kg bw (calculated to be between 48.2 and 55.7 mg/kg bw) for three consecutive days. Extracts from tissues, urine, feces and stomach contents in this study were analyzed by liquid scintillation counting (LSC) and thin layer chromatography (TLC). For metabolite characterization, the methanol extracts of male and female rat liver samples were examined, along with urine samples collected after the start of dosing from one male and one female animal (4 hours after first dosing). For the 9-hour animals, greater than 68% of the total [^{14}C]-residue in liver was extracted with methanol. For the 15-hour animals, the [^{14}C]-residues extracted with methanol decreased to an average of 48%. Additional extractions and analysis of rat kidney and stomach content samples, as well as 48-hour feces samples, were carried out for the 9-hour rats. Greater than 90% of the total [^{14}C]-residue present in these samples was extracted with methanol.

Tissue extracts from the total residue studies in cattle and swine were analyzed for metabolites. Overall, the metabolites found in rat liver samples and urine correspond to those found in liver and urine of cattle and swine. The exception is that 2-pyrrolidone was found in cattle, but not in any rat samples examined. This difference may be due to rats being sacrificed at 9 and 15 hours after dosing while cattle were slaughtered 24 hours after dosing.

Table III.4. Percentage of total residues recovered from tissues in rats administered by oral gavage [^{14}C]-NMP at a nominal rate of 50 mg/kg bw for three consecutive days.

Tissue	Time of Sacrifice (hours)	Animal ID	% total residue in methanol extract	% total residue in post extracted solids	Total % total residue recovered
Liver	9	1 + 2	81.7	17.8	99.5
Liver	9	9 + 6	68.8	25.5	94.3
Liver	15	9 + 5	48.9	43.7	92.6
Liver	15	1 + 3	47.4	44.6	92.0
Kidney	9	1 + 2	92.8	7.57	100.4
Kidney	9	9 + 6	92.1	9.07	101.2
Stomach Contents	9	1 + 2	102.1	1.63	103.7
Stomach Contents	9	9 + 6	101.6	1.08	102.7
Feces collected 48 hours after initial dosing	9	1 + 2	91.6	9.16	100.8
Feces collected 48 hours after initial dosing	9	9 + 6	96.4	3.06	99.5

Table III.5. Percentage of total residues recovered from livers of cattle and swine administered a single intramuscular injection of 41.32 mg [^{14}C]-NMP/kg bw.

Animal Tissue	Time of Sacrifice (days)	Animal ID	% total residue in methanol extract	% total residue in post extracted solids	Total % total residue recovered
Cattle Liver	1	9177	87.9	3.95	91.9
Cattle Liver	1	9217A	91.6	4.48	96.1
Swine Liver	1	7	94.8	8.48	103.3
Swine Liver	1	1	92.4	11.4	103.8

D. Tolerance and R_m for Residues

The tolerances for residues established for the RLNAD product apply to the generic product. The tolerances for the sum of residues of the tetracyclines including chlortetracycline, oxytetracycline, and tetracycline are 2 ppm in cattle and swine muscle, 6 ppm in cattle and swine liver, 12 ppm in cattle and swine fat, and 12 ppm in cattle and swine kidney (21 CFR 556.500).

R_m is the concentration of the marker residue in the target tissue when the residue of carcinogenic concern is equal to S_m. The R_m for carcinogenic residues, established for parent NMP, is 180 ppm in cattle liver and 88 ppm in swine liver.

E. Withdrawal Period

The withdrawal period is that previously assigned to the RLNAD product. The withdrawal period for oxytetracycline injection is 28 days for beef cattle, non-lactating dairy cattle, and swine (21 CFR 522.1660a). Total residues of NMP are below the S_m values at 1-day withdrawal, therefore the withdrawal period for Oxytetracycline Injection is determined by the depletion of oxytetracycline.

F. Analytical Method for Residues

The regulatory analytical method for detection of residues of oxytetracycline is a cylinder plate diffusion microbiological assay using *Bacillus cereus* var. *mycoides* (ATCC 11778). The method is published by the Food and Drug Administration, "Antibiotic Residues in Milk, Dairy Products and Animal Tissues: Methods, Reports, and Protocols", Revised October 1968, reprinted December 1974.

The validated analytical method for analysis of residues of oxytetracycline is on file at the Center for Veterinary Medicine, 7500 Standish Place, Rockville, MD 20855. To obtain a copy of the analytical method, please submit a Freedom of Information request to: https://www.accessdata.fda.gov/scripts/foi/FOIRequest/requestinfo.cfm.

The animal drug regulations at 21 CFR 500.1410 provide for the incorporation by reference of the validated regulatory method for NMP. To obtain a copy of the analytical method, please submit a Freedom of Information request to: https://www.accessdata.fda.gov/scripts/foi/FOIRequest/requestinfo.cfm. Alternatively, a copy of the method may be inspected at the Office of the Dockets Management Staff (HFA-305), Food and Drug Administration, 5630 Fishers Lane, Rm. 1061, Rockville, MD 20852, (301) 827-6860, between 9 a.m. and 4 p.m., Monday through Friday or at the National Archives and Records Administration (NARA). For information on the availability of this material at NARA, call (202) 741-6030, or go to: http://www.archives.gov/federal-register/cfr/ibr-locations.html.

IV. USER SAFETY

N-methyl-2-pyrrolidone (NMP), an ingredient in Oxytetracycline Injection, has been reported to cause reproductive and developmental toxicities in laboratory animals following high, repeated exposures, as described in the following documents from FDA and two other regulatory agencies:

1. FDA/CVM: The FOI Summary for the original approval of ANADA 200-117, dated April 13, 1995.

2. European Chemical Agency (ECHA): How to comply with REACH Restriction 71, guideline for users of NMP (1-methyl-2-pyrrolidone). ECHA Reference #: ECHA-19-H-07-EN July 2019 [https://echa.europa.eu/-/advice-on-how-to-comply-with-nmp-restriction].

3. United States Environmental Protection Agency (U.S. EPA): Risk Evaluation for n-Methylpyrrolidone (2-Pyrrolidinone, 1-Methyl-) (NMP). EPA Document# EPA-740-R1-8009 December 2020, conducted by the Office of Chemical Safety and Pollution Prevention, Office of Pollution Prevention and Toxics (OPPT) [https://www.epa.gov/sites/production/files/2020-12/documents/1_risk_evaluation_for_n-methylpyrrolidone_nmp_casrn_872-50-4.pdf]

Because persons who are pregnant that handle Oxytetracycline Injection may be exposed to NMP via accidental injection or dermally, the Agency recommends that pregnant women wear gloves and exercise caution when handling Oxytetracycline Injection, or avoid handling the product.

The product labeling contains the following information regarding safety to humans handling, administering, or exposed to Oxytetracycline Injection:

USER SAFETY WARNINGS:
Not for use in humans. Keep out of reach of children.
Reproductive and developmental toxicities have been reported in laboratory animals following high, repeated exposures to NMP. Pregnant women should wear gloves and exercise caution or avoid handling this product. To obtain a Safety Data Sheet (SDS), contact Bimeda, Inc. at 1-888-524-6332.

V. AGENCY CONCLUSIONS

The information submitted in support of this supplemental ANADA satisfy the requirements of section 512(c)(2) of the Federal Food, Drug, and Cosmetic Act. The data demonstrate that Oxytetracycline Injection, when used according to the label, is safe and effective for the indications listed in Section I.M. above.

Additionally, data demonstrate that residues in food products derived from beef cattle, non-lactating dairy cattle, and swine treated with Oxytetracycline Injection will not represent a public health concern when the product is used according to the label.

Labeling restricts this drug to use by or on the order of a licensed veterinarian. This decision aligns the labeling with the referenced listed new animal drug and was based on the following factors: (a) adequate directions cannot be written to enable lay persons to appropriately diagnose and subsequently use this product for the labeled indications and (b) restricting this drug to use by or on the order of a licensed veterinarian should help prevent indiscriminate use which could result in violative tissue residues.

VI. APPENDIX

Original text:

The R_m for carcinogenic residues, established for parent NMP, is 700 ppb in cattle liver and 500 ppb in swine liver.

Revised text (January 3, 2024):

The R_m for carcinogenic residues, established for parent NMP, is 180 ppm in cattle liver

and 88 ppm in swine liver.