

I. GENERAL INFORMATION

A. File Number

NADA 138-992

B. Sponsor

The Upjohn Company, Agricultural Division

C. Proprietary Name

MGA[®]100/200 Premix/MGA[®]500 Liquid Premix
Bovatec[®]
Tylan[®]

D. Established Name

melengestrol acetate
lasalocid sodium
tylosin phosphate

E. Dosage Form

Feed

F. Dispensing Status

OTC

G. Dosage Regimen

melengestrol acetate - 0.25 to 0.5 mg/head/day
lasalocid sodium - 10 to 30 g/ton of air dried complete feed
tylosin phosphate - 8 to 10 g/ton of air dried complete feed (to provide 90 mg/hd/day)

Approval has been granted to feed 0.25 to 0.5 mg melengestrol acetate (MGA) per head per day in combination with 10 to 30 g lasalocid per ton of air dried complete feed and 8 to 10 g tylosin per ton of air dried complete feed (to provide 90 mg/hd/day) when each additive is provided via separate supplements or a supplement containing melengestrol acetate is fed in combination with a complete feed containing lasalocid and tylosin.

The supplement containing melengestrol acetate provided at a rate of 0.5 to 2.0 pounds per head may be top-dressed onto or mixed with the feed containing monensin and tylosin.

This clearance does not provide for the mixing of melengestrol acetate, monensin and tylosin premixes together in a common supplement or in a complete ration offered for sale by the feed manufacture.

H. Route of Administration

Oral

I. Indication

For increased rate of weight gain, improved feed efficiency, suppression of estrus (heat), and reduced incidence of liver abscesses in heifers fed in confinement for slaughter.

II. EFFECTIVENESS (In compliance with combination approval claims)

The efficacy of melengestrol acetate for increased rate of weight gain, improved feed efficiency and suppression of estrus (heat) in heifers fed in confinement for slaughter is well documented and approved (21 CFR 558.342). Further, the effectiveness of lasalocid for improved feed efficiency and increased rate of weight gain of feedlot cattle is established and approved (21 CFR 558.311) as is the effectiveness of tylosin for reduction of liver abscesses in the same class of cattle (21 CFR 558.625). For these applications, the efficacy of melengestrol acetate, lasalocid and tylosin when co-administered was established by conducting a series of field trials under a common protocol. These trials were designed to evaluate the performance of feedlot heifers fed each drug separately and in combination.

A. Pivotal Study

A feedlot heifer study consisting of field trials at nine locations with three replications per location was conducted to generate animal performance data on the combination of melengestrol acetate, lasalocid and tylosin. Results of each study are reported as well as a summary and statistical analysis of the pooled data from all nine field investigations.

1. Type of Study
 - a. Clinical (field) trials
2. Investigator names and addresses by location:

Trial No. 540-9665-0-JFM-83-002

Steve Rust, Ph.D.
Montana Ag Experiment Station
Route 1, Box 131
Huntley, MT 59037

Trial No. 540-9665-0-JFM-83-003

David Bechtol, D.V.M.
Palo Duro Agri. Division
Box 974
Canyon, TX 79015

Trial No. 540-9665-0-JFM-83-004

Thomas R. Schriemer, B.S.
Unit 9690
The Upjohn Company
Kalamazoo, MI 49001

Trial No. 540-9665-0-JFM-83-005

Steve Rust, Ph.D.
Montana State University
Bozeman, MT 59717

Trial No. 540-9665-0-JFM-83-006

Jack Riley, Ph.D.
Dept. of Animal Science - Industry
Weber Hall KSU
Manhattan, KS 66506

Trial No. 540-9665-0-JFM-83-007

Richard Luther, Ph.D.
Animal and Range Science Dept.
Animal Science Complex Building
Brookings, SD 57007

Trial No. 540-9665-0-JFM-83-008

Jerry Martin, Ph.D.
Panhandle State University
Box 430
Goodwell, OK 73939
- and -
Donald R. Gill, Ph.D.
005 Animal Science
Oklahoma State University
Stillwater, OK 74074

Trial No. 540-9665-0-JFM-83-009*

Elvin Thomas, Ph.D.
Dept. of Animal & Dairy Sciences
Auburn University
Auburn, AL 36830

Trial No. 540-9665-0-JFM-83-010

Wilton Heinemann, Ph.D.
Research & Extension Center
Washington State University
P.O. Box 30
Prosser, WA 99350

Trial No. 540-9665-0-JFM-84-001

Steve Schmidt, Ph.D.
Dept. of Animal and Dairy Sciences
Auburn University
Auburn, AL 36830

*This trial was canceled shortly after it was started for reasons unrelated to the safety and efficacy of the compounds. There were no data generated as a result of this trial.

3. General design of each field investigation

- a. Purpose of the field investigation: To evaluate the effect of melengestrol acetate, lasalocid sodium and tylosin phosphate on the performance of feedlot heifers when fed singly and in combination.
- b. Test animals
 - (i) Species: Bovine
Number/group: The number of heifers per treatment group varied from location to location, and ranged from 6 to 14 animals per pen (Table 1).
 - (ii) Subgroup Identity: Heifers used in the field studies were non-pregnant yearlings and the breeds were typical of those commonly found in commercial feedlots. The average initial weights ranged from 674 to 826 pounds (Table 1).
- c. Control groups

Groups of heifers fed melengestrol acetate, or lasalocid or tylosin alone, or lasalocid and tylosin in combination served as controls. Pairwise, comparisons were made to groups of heifers for the 3 drugs in combination.
- d. Diagnosis: Not applicable.
- e. Dosage form
 - (i) Medicated feed: Feed was medicated using melengestrol acetate, lasalocid or tylosin premixes. Final feeds were prepared using intermediate premixes which were either mixed into or top-dressed onto the bulk of the ration.
- f. Route of drug administration:
 - (i) Oral
- g. Dosages: The dosages used in the field study were: melengestrol acetate, 0.5 mg/head/day; lasalocid, 30 gm/ton of air dried feed; tylosin, 10 gm/ton of air dried feed.
- h. Days on test: The cattle were fed for a minimum of 90 days, the actual day of trial termination being left up to the investigator's judgement as to when the cattle had reached market weight and condition. The days on test by location are listed on Table 1.

Table 1. General Trial Information

Location Trial No.	Number/Pen	Mean Starting Wt/lbs	Days on Test	Estrus Detection (Yes/No)
Montana (Huntley) (A) 540-9665-0-JFM-83-002	12	790	102	Yes
Texas 540-9665-0-JFM-83-003	14	707	104	Yes
Michigan 540-9665-0-JFM-83-004	8	735	97	No
Montana (Bozeman) (B) 540-9665-0-JFM-83-005	8	826	97	Yes
Kansas 540-9665-0-JFM-83-006	6	731	92	No
South Dakota 540-9665-0-JFM-83-007	8	700	126	No
Oklahoma 540-9665-0-JFM-83-008	8	674	112	Yes
Washington 540-9665-0-JFM-83-010	8	773	96	Yes
Alabama 540-9665-0-JFM-84-001	8	728	99	No

i. Measurements taken: The following were measured:

Estrus: 2X per day (AM and PM) during days 5 through 47 of the trial. Estrus was measured at five of the nine locations (Table 1).

Cattle weight: cattle were weighed at about 28 day intervals.

Feed consumption: daily feed records were maintained.

Daily observations: individual pens of cattle were observed at least once per day for general activity and physical condition of the cattle.

Liver Evaluation: individual heifer livers were evaluated for liver abscesses at slaughter.

4. Results

Performance Data: The treatment by location means for percent standing estrus, average daily gain, feed efficiency and percent abscessed livers are presented in the following tables. While the study included several treatment groups, data from only those pertinent to the subject approval are included.

Table 2. Treatment by Location Means for Percent Standing Estrus

Treatment	Oklahoma	Montana*	Montana**	Texas	Washington
melengestrol acetate/tylosin	8.33	16.67	16.67	26.19	8.33
melengestrol acetate/lasalocid sodium	8.33	16.67	11.11	11.90	16.67
lasalocid sodium/tylosin	45.83	36.31	61.11	19.05	37.50
melengestrol acetate/lasalocid sodium/tylosin	4.17	16.67	8.33	7.14	8.33

*Bozeman, Montana

**Huntley, Montana

Table 3. Treatment by Location Means for Average Daily Gain

Treatment	Alabama	Oklahoma	Kansas	Montana*	Montana**	S. Dakota	Texas	Michigan	Washington
melengestrol acetate/tylosin	2.63	2.39	3.17	2.40	2.93	3.19	2.92	3.42	2.52
melengestrol acetate/lasalocid sodium	3.05	2.38	3.09	2.43	2.87	3.31	3.15	3.45	2.59
lasalocid sodium/tylosin	2.91	2.10	2.99	2.44	2.60	3.05	2.61	3.04	2.36
melengestrol acetate/lasalocid sodium/tylosin	3.03	2.28	3.21	2.61	3.00	3.32	2.89	3.42	2.78

*Bozeman, Montana

**Huntley, Montana

Table 4. Treatment by Location Means for Feed Efficiency

Treatment	Alabama	Oklahoma	Kansas	Montana *	Montana **	S. Dakota	Texas	Michigan	Washington
melengestrol acetate/tylosin	8.53	6.25	8.19	10.42	9.03	6.53	7.49	6.85	8.49
melengestrol acetate/lasalocid sodium	7.47	6.19	8.02	10.07	9.14	9.56	7.26	6.93	8.17
lasalocid sodium/tylosin	7.68	6.79	8.36	10.43	9.67	6.78	8.29	7.29	8.60
melengestrol acetate/lasalocid sodium/tylosin	7.49	6.43	7.74	9.84	8.88	6.37	7.63	6.77	7.83

*Bozeman, Montana

**Huntley, Montana

Table 5. Treatment by Location Means for Percent Abscessed Livers

Treatment	Alabama	Oklahoma	Kansas	Montana *	Montana **	S. Dakota	Michigan	Washington
melengestrol acetate/tylosin	12.50	0.00	0.00	8.33	11.11	4.17	0.00	8.33
melengestrol acetate/lasalocid sodium	29.17	16.67	33.33	54.17	19.44	8.33	8.33	12.50
lasalocid sodium/tylosin	8.33	0.00	5.56	13.69	8.33	0.00	0.00	26.19
melengestrol acetate/lasalocid sodium/tylosin	16.67	0.00	5.56	25.00	19.44	0.00	0.00	12.50

*Bozeman, Montana

**Huntley, Montana

The results from the 9 individual field investigations were pooled and analyzed statistically to evaluate effectiveness. Average daily gain (ADG), feed efficiency (FE) (9 locations), and liver abscess data (8 locations) were pooled. Estrus suppression data were pooled from five of the nine locations.

The least squares treatment means pooled across all locations are presented in Table 6.

Table 6. Least Squares Treatment Means Pooled Across Location

Treatment	% Standing Estrus*	Feed Efficiency	Average Daily Gain	% Abscessed Livers*
melengestrol acetate/tylosin	23.89	7.97	2.84	16.19
melengestrol acetate/lasalocid sodium	22.41	7.76	2.92	27.94
lasalocid sodium/tylosin	39.66	8.21	2.68	17.39
melengestrol acetate/lasalocid sodium/tylosin	17.98	7.66	2.95	17.76

*Means are based on Freeman-Tukey transformation

5. Statistical analysis:

A least squares analysis of variance for the transformed liver abscess (8 locations) and performance data from the nine location, and transformed estrus data from 5 locations was conducted. The results of this analysis are presented on Table 7.

Table 7. The significance levels for the comparisons required for the combinations were:

Combination	Comparison	Parameter			
		Estrus	FE	ADG	Liver Abscess
MGA + Lasalocid (L) + T	MGA + L vs MGA + L + T				0.00021
	MGA + T vs MGA + L + T		0.00531	0.02985	
	L + T vs MGA + L + T	0.000012	0.00001	0.000005	

The melengestrol acetate + lasalocid + tylosin combination was justified since the contribution of lasalocid significantly enhanced feed efficiency (P=0.00531). In this combination, melengestrol acetate was effective for estrus suppression (P=0.000012), feed efficiency (P=0.00001), and average daily gain (P=0.000005). Tylosin was also effective in reducing the incidence of liver abscesses (P=0.00021) in this 3-way combination (Table 7).

6. Conclusions

The data generated by the nine location field study demonstrated that the melengestrol acetate, lasalocid, tylosin combination is justified for its effect on estrus suppression, improved feed efficiency, increased rate of weight gain and reduction of liver abscesses. Each drug, melengestrol acetate, lasalocid and tylosin has been shown to make a significant contribution to the effectiveness of the combination.

7. Adverse Reactions

No adverse reactions due to the feeding of melengestrol acetate, lasalocid, or tylosin, either singly or in combination were reported.

8. Special Issues

Compliance with combination drug policy:

- a. The effectiveness and safety of melengestrol acetate, lasalocid, and tylosin when fed singly to feedlot cattle are well documented in their respective approved NADAs. The results of effectiveness and safety studies were submitted to the FDA in support of these respective NADAs. Such data resulted in the approval of melengestrol acetate as per CFR 558.342, lasalocid as per CFR 558.311, and tylosin as per CFR 558.625. Data generated in support of combination usage have demonstrated that

melengestrol acetate, lasalocid and tylosin fed together are effective and safe.

The data establish compliance with 21 CFR 514.1(b)(8)(v) and CVM's combination drug guidelines (November 9, 1983). The combination is justified as detailed in items 4, 5 and 6 above.

The use of untreated controls was not deemed necessary as a test for general effectiveness because the individual drugs have been previously shown to be effective and already approved at the levels tested. The treatment groups consisting of the individual drugs and the two-way combinations served as the appropriate controls for the respective three-way combination. Thus, these treatment groups essentially served as negative controls with respect to 21CFR 514.111 (a)(5)(ii)(a)(4), and the studies were properly controlled.

To justify the combination, comparisons were made which demonstrated that each drug made a statistically significant contribution to the combination. The three-way combination provides a benefit that cannot be obtained from any of the possible two-way combinations.

A comparison of melengestrol acetate-lasalocid-tylosin vs. melengestrol acetate-lasalocid for transformed percent liver abscesses demonstrates the contribution of tylosin to the combination.

A comparison of melengestrol acetate-lasalocid-tylosin vs. lasalocid-tylosin for ADG, FE and estrus suppression demonstrates the contribution of melengestrol acetate to the combination. Significance for any variable shows that melengestrol acetate contributes to the combination.

A comparison of melengestrol acetate-lasalocid-tylosin vs. melengestrol acetate-tylosin for ADG and FE demonstrates the contribution of lasalocid to the combination. Significance for any variable shows that lasalocid contributes to the combination.

The proposed ranges for each drug are justified when fed in combination since the highest levels proposed were tested in combination for efficiency and human and animal safety.

B. Corroborative Studies: N/A

III. TARGET ANIMAL SAFETY

A. Pivotal study

The safety of melengestrol acetate, lasalocid and of tylosin in the bovine have been documented as evidenced by their respective approvals (21 CFR 558.342 for melengestrol acetate, 21 CFR 558.311 for lasalocid, and 21 CFR 558.625 for tylosin) as feed additives. The following pivotal study was conducted to establish the target animal safety of these three additives when fed in combination.

1. A 90 day, subchronic study was conducted.
2. Investigator: A.D. Hall, Ph.D., D.V.M.
The Upjohn Company
Kalamazoo, MI 49001
3. General Study design:
 - a. Purpose: The purpose of this study was to evaluate the safety of the melengestrol acetate, lasalocid and tylosin when fed in combination in the diet of yearling heifers at 1 to 5 times the recommended dosage.
 - b. Test animals:
 - (i) Species: Bovine
Number per Group: 14 control, 7 treated.
 - (ii) Breed: Hereford
Age: Yearling
Sex: Female (heifers)
Weight: 284 to 378 kg initial weight
 - c. Dosage form:
 - (i) Drug premixes were incorporated in the feed through the use of intermediate premixes.
 - (ii) Formulations were typical of those used in commercial feedlot.
 - d. Dosage used:
 - (i) Control: No drugs
 - (ii) 1X: 0.5 mg melengestrol acetate per head per day
30 g lasalocid sodium per ton of air dried feed
10 g tylosin phosphate per ton of air dried feed
 - (iii) 3X: 1.5 mg melengestrol acetate per head per day
90 g lasalocid sodium per ton of air dried feed
30 g tylosin phosphate per ton of air dried feed
 - (iv) 5X: 2.5 mg melengestrol acetate per head per day
150 g lasalocid sodium per ton of air dried feed
50 g tylosin phosphate per ton of air dried feed
 - e. Routes of administration: Oral
 - f. Test duration: 90 days
 - g. Parameters measured:
 - (i) Clinical observations included twice daily health checks, daily observations, body weight, daily feed consumption.

(ii) Clinical pathology

The following hematology parameters were measured:

- Total leukocyte count
- Total erythrocyte count
- Hemoglobin
- Hematocrit
- Mean corpuscular volume
- Mean corpuscular hemoglobin
- Mean corpuscular hemoglobin concentration
- Platelets

Microscopic Examination: WBC differential

The following serum chemistry parameters were measured:

- Aspartate aminotransferase
- Alkaline phosphatase
- Creatinine
- Inorganic Phosphorus
- Glucose
- Blood Urea Nitrogen
- Calcium
- Cholesterol
- Total Bilirubin
- Total Protein
- Albumin
- Creatinine phosphokinase
- Sodium
- Potassium
- Chloride
- Sorbitol dehydrogenase

Urine specimens were collected via paracentesis from the urinary bladder at the time of necropsy. The following parameters were measured:

- Specific gravity
- Dipstick:
 - pH
 - Protein
 - Glucose
 - Ketones
 - Bile

Microscopic Examination: Formed elements

(iii) Gross and microscopic pathology

The following is a list of organs and tissues that were examined grossly and had a representative sample fixed in 10% neutral buffered formalin for microscopic evaluation:

Brain
Pituitary
Thyroids
Adrenals
Pancreas
Ovaries
Mediastinal lymph node
Mesenteric lymph node
Lung
Liver
Kidneys
Urinary bladder
Spleen
Bone Marrow (smear)*
Gross lesions
Diaphragm
Rumen
Reticulum
Omasum
Abomasum
Duodenum
Cecum
Colon
Heart (one section from each atrium and ventricle)
Gall bladder
Aorta (and small arteries)
Bone

*Bone marrow smears were made at the time of necropsy, air dried and fixed for 30 seconds or more in methanol. Slides were stained with Wrights stain.

The following organs were weighed at necropsy, paired organs weighed together:

Adrenals, liver, kidneys and heart.

(iv) Results

(a) Clinical Observations

No adverse, drug-related clinical signs were observed during this study. Although there were no significant differences in average daily gain (ADG) between treated and control animals at mid-study and at termination, there was a trend toward decreased ADG at the 3X and 5X levels of treatment.

(b) Clinical pathology

Occasional statistically significant changes in several hematologic and serum chemistry parameters were detected, but all values were well within normal acceptable ranges and no dose-related trends were seen.

(c) Gross and microscopic pathology

Several gross and histologic tissue changes were observed, but due to the relative even distribution of the changes in all dose groups including controls, these findings were considered to be incidental rather than drug-related.

(v) Conclusions

Based upon the results of this study, the combination of melengestrol acetate, lasalocid and tylosin phosphate in the daily feed of yearling heifers appears to be non-toxic when fed for 90 days at up to 5 times the recommended dose.

IV. HUMAN FOOD SAFETY

A. Drugs for use in food animals

1. Toxicity tests

Data regarding toxicity testing in melengestrol acetate, lasalocid sodium and tylosin phosphate are contained in the approved NADAs for the three above mentioned compounds.

2. Safe concentration of residues

Tolerances for melengestrol acetate, lasalocid sodium and tylosin phosphate are published in the Code of Federal Regulations. Melengestrol acetate is currently approved under 21 CFR 558.342 for use in heifers at 0.25 to 0.50 mg/head/day with a 48 hour withdrawal period. No residues of melengestrol acetate may be found in uncooked edible tissues of cattle with a method sensitive to 25 ppb (21 CFR 556.380). The tolerance for lasalocid in edible tissue of cattle under 21 CFR 556.347 is 0.7 ppm with no withdrawal period established for cattle under 21 CFR 558.311. The tolerance for tylosin phosphate has been established at 0.2 ppm (negligible residue) in uncooked fat, muscle, liver and kidney (CFR 556.740). Tylosin phosphate has no withdrawal period established under 21 CFR 558.625.

3. Residue depletion and metabolism studies

Numerous studies have been conducted relative to the metabolism and depletion of residues of melengestrol acetate, lasalocid, and tylosin when administered individually to cattle. The results of those studies have been filed under the following submissions:

<u>Melengestrol Acetate</u>	<u>Lasalocid</u>	<u>Tylosin</u>
NADA 34-254	NADA 96-298	NADA 12-491
NADA 39-402		NADA 104-646
NADA 124-309		

4. Residue depletion noninterference study

The following study was conducted to support the use of the three-way combination of melengestrol acetate, lasalocid sodium and tylosin phosphate in heifers:

Location: Michigan

Sponsor: The Upjohn Company

Investigators: L.F. Krzeminski, Ph.D.
The Upjohn Company

P. R. Handy, Ph.D.
Lilly Research Laboratories
Greenfield, IN 46140
G. Chen, Ph.D.
Animal Science Research
Hoffman-LaRoche
Nutley, NJ 07110

Groups of feedlot heifers were fed for 90 days with the drug combinations and treatment levels described below.

Group	Number of Animals	Treatment Level	Treatment*
1	14	0	Control
2	7	1X	0.5 mg MGA + 30 g lasalocid + 10 g tylosin
3	7	3X	1.5 mg MGA + 90 g lasalocid + 30 g tylosin
4	7	5X	2.5 mg MGA + 150 g lasalocid + 50 g tylosin

*Melengestrol acetate treatment expressed as mg/head/day, lasalocid and tylosin expressed as gram/ton air dried ration.

The animals in each group were slaughtered within 16 hours following their last feeding. Perirenal fat was collected from all four treatment groups for the melengestrol acetate analysis, and liver samples were collected from the group 1 and 2 animals for the assay of lasalocid and tylosin.

a. Residues of melengestrol acetate

The samples of perirenal fat from each animals were assayed for residues of melengestrol acetate using the official AOAC gas chromatographic method. Analysis of the samples from the group 2 (1X) animals showed that all fat samples were below the 25 ppb tolerance for MGA and five of the seven samples were below the 10 ppb limit of reliable measurement of the assay. The two fat samples that gave positive responses had MGA levels of 12.7 and 13.7 ppb.

Fat samples from groups 3 and 4 (the heifers dosed at 3X and 5X levels) yielded average residue levels of 37.6 and 49.4 ppm, respectively.

b. Residues of lasalocid

Liver tissue that was collected from the seven control animals and the seven heifers fed the group 2 (1X) ration were assayed for lasalocid by the hplc-fluorescence regulatory method for the drug. An average residue value of 0.09 ppm (SD. 0.11 ppm) was obtained for the group 2 heifers, and that level is well below the 0.7 ppm tolerance for lasalocid in the livers from the control animals.

c. Residues of tylosin

The liver tissue samples from the group 2 (1X) heifers and seven of the control animals were assayed for tylosin by the official assay which measures microbiologically the active drug. No positive responses were obtained for any of the samples by this method which has a limit of quantitation of 0.1 ppm.

5. Assay noninterference data

a. Melengestrol acetate assay

Data were generated using spiked control fat tissue samples to demonstrate that the presence of residues of lasalocid and tylosin does not interfere with the assay for MGA. The study was also intended to show that MGA is stable in frozen fat tissue over a period of 60 days.

Samples of freshly ground control bovine fat tissue were spiked with MGA, lasalocid and tylosin at the following levels:

MGA: 0.025
 Lasalocid sodium: 7.0 ppm
 Tylosin phosphate: 1.0 ppm

The spiked samples were assayed in duplicate for MGA by the official AOAC method after 0, 15, 30, 45, and 60 days of storage at -20°C. The average recoveries are shown below.

Recovery of MGA from Bovine Fat

	Day of Assay						Ave.	S.D.
	0	15	30	45	60			
ppb found	24.1	20.8	24.5	25.3	24.4	23.8		
Percent Recovery	96	83	98	101	98	95	7.0	

b. Lasalocid assay

Data demonstrating the noninterference of residues of melengestrol acetate on the HPLC-fluorescence detection assay for lasalocid were generated by Hoffmann-LaRoche to support the original NADA approval for lasalocid in cattle (NADA 96-298). Those data have also been

published as a journal article (J. Agricultural and Food Chemistry, 31, 75-78 (1983)) and are summarized below.

Recovery Values Obtained for Lasalocid from Spiked Control Liver Samples

Drug Combination and Fortification Level	Lasalocid Recovery
Lasalocid (0.1 ppm)	86% (mean of 6 assays)
Lasalocid (0.1 ppm) + tylosin (0.40 ppm)	85.0%
Lasalocid (0.1 ppm) + MGA (0.11 ppm)	80.1%

c. Tylosin assay

Data to demonstrate that the presence of lasalocid and MGA does not interfere with the assay of tylosin are contained in study S-AAC-84-03 conducted by the Elanco Products Company. Samples of liver from control heifers were composited and fortified with 0.2 ppm lasalocid, 0.2 ppm tylosin and 0.2 ppm MGA. The samples were assayed for tylosin by the cylinder plate microbiological method using *Micrococcus luteus* as the test organism. No interference was detected by the presence of lasalocid and MGA.

The stability of tylosin in frozen liver tissue was demonstrated in study MRC8910 conducted by the Elanco Products Company. Liver tissue from control animals was fortified with 0.2 ppm tylosin and then frozen. Portions of the samples were thawed at intervals up to 56 days and assayed for tylosin. The stability of tylosin through 56 days was indicated by the recoveries in the following table:

Tylosin Storage Stability in Frozen Liver

Storage (days)	Mean Tylosin Recovery (ppm)	Percent Theoretical
0	0.166	83%
14	0.239	114%
28	0.172	86%
56	0.162	81%

Regression analysis of these data demonstrates satisfactory tylosin stability for the period the tissues in the residue depletion study were stored before assay.

The residue depletion and assay non-interference studies presented above demonstrate that the combined feeding of melengestrol acetate, lasalocid sodium, and tylosin phosphate at their highest approved levels results in tissue residues below the tolerance levels for each of the three drugs at 16 hours of withdrawal. The data also show that each drug in the three-way combination does not interfere with the assays for the others. This work confirms the adequacy of the 48-hour withdrawal time required for the presence of MGA in the combination and demonstrates that the use of these feed additives in combination does not pose a hazard to public health.

6. Regulatory methods

Practical analytical methods of analysis for tissue residues of melengestrol acetate, lasalocid sodium and tylosin phosphate may be found in the Food Additives Analytical Manual on display in FDA's Freedom of Information Public Room (Room 12-A-30), 5600 Fishers Lane, Rockville, MD 20857.

V. AGENCY CONCLUSIONS

These NADAs provide for the combination use of MGA, lasalocid sodium, and tylosin phosphate at the levels of 0.25 to 0.5 mg/head/day, 10 to 30 g/ton of feed, and 8 to 10 g/ton of feed (to supply 90 mg/head/day), respectively; for increased rate of weight gain, improved feed efficiency, suppression of estrus (heat), and reduced incidence of liver abscesses in heifers fed in confinement for slaughter. Adequate data were submitted which show that the MGA, lasalocid, tylosin combination is justified for its effect on estrus suppression, improved feed efficiency, increased rate of weight gain and reduction of liver abscesses. Each drug; MGA, lasalocid, and tylosin has been shown to make a significant ($P < 0.05$) contribution to the effectiveness of the combination. The combination, when fed at up to 5X the highest recommended combination approval (0.5 mg MGA/head/day plus 30 g lasalocid/ton, and 10 g tylosin phosphate/ton for 90 days), did not produce an adverse effect. No changes were made in the approved levels of the compounds or in the target animal and the noninterference of tylosin phosphate, lasalocid sodium and MGA with the analytical methods for MGA, lasalocid sodium and tylosin phosphate, respectively, was demonstrated. Accordingly, approval of these NADAs is not expected to increase human exposure to drug residues, and therefore did not require a complete re-evaluation of the human safety data in the original applications. For the purposes of human food safety review, these original NADAs have been treated as Category II supplements under the Agency's Supplemental Policy (42 FR 64367). These production drugs are OTC because they do not raise any special safety concerns.

VI. ATTACHMENTS

Type A Medicated Feed package label and mixing instructions
Type B Medicated Feed package label and mixing instructions
Type C Medicated Feed package label and mixing instructions

Copies of these labels may be obtained by writing to the:

Food and Drug Administration
Freedom of Information Staff (HFI-35)
5600 Fishers Lane
Rockville, MD 20857

Or requests may be sent via fax to: (301) 443-1726. If there are problems sending a fax, call (301) 443-2414.

The format of this FOI Summary document has been modified from its original form to conform with Section 508 of the Rehabilitation Act (29 U.S.C. 794d). The content of this document has not changed.