**Hydrolysis of Pymetrozine**

**Report:** McDonald, J. 1996. Supplemental report hydrolysis of $^{14}$C-[triazine-6]CGA-215944 under laboratory conditions at pH 5. Laboratory Project ID: ABR-96020. Study No. 85-96. Unpublished study performed and submitted by Ciba-Geigy Corporation, Greensboro, NC.

Kirkpatrick, D. 1995. The hydrolysis of CGA-215944 under laboratory conditions. Laboratory Project ID: CBG 570. Unpublished study performed by Huntington Life Sciences Ltd., Cambridgeshire, ENGLAND; and submitted by Ciba-Geigy Limited, Postfach, Switzerland.

**Document No.:** MRID 44024962 and MRID 44024963

**Guideline:** OPPTS 835.2120

**Classification:** A study classification was not provided on the attached Data Evaluation Record, therefore, these studies are now classified as **acceptable**. Together, these studies are scientifically valid and provide useful information on the hydrolysis of pymetrozine.

**PC Code:** 101103

**Reviewers:** Jessica L. O. Joyce, Fate Scientist
U.S. EPA Date: Date: 2018.02.08 07:29:58 -05'00'

Rochelle F.H. Bohaty, Senior Scientist
U.S. EPA Date: 2018.02.08 07:17:36 -05'00'

TEXT SEARCHABLE DOCUMENT 2011

# DATA EVALUATION RECORD

## STUDY 1a

CHEM 101103 Pymetrozine §161-1
CAS No. 123312-89-0
FORMULATION--00--ACTIVE INGREDIENT

STUDY ID 44024963
Kirkpatrick, D. 1995. The hydrolysis of CGA-215944 under laboratory conditions. Laboratory Project ID: CBG 570. Unpublished study performed by Huntington Life Sciences Ltd., Cambridgeshire, ENGLAND; and submitted by Ciba-Geigy Limited, Postfach, SWITZERLAND.

STUDY ID 44024962
McDonald, J. 1996. Supplemental report: hydrolysis of $^{14}$C-[triazine-6]CGA-215944 under laboratory conditions at pH 5. Laboratory Project ID: ABR-96020. Study No. 85-96. Unpublished study performed and submitted by Ciba-Geigy Corporation, Greensboro, NC.

DIRECT REVIEW TIME = 60 hours

REVIEWED BY: D. A. Saccone, B.S.
Signature:
TITLE: Scientist
Date:

EDITED BY: P. C. DeLeo, Ph.D.
Signature:
TITLE: Senior Scientist Date:

APPROVED BY: P. H. Howard, Ph.D.
Signature:
TITLE: Project Manager
Date:

ORG: Syracuse Research Corp.
Arlington, VA 22202

TEL: 703/413-9369

APPROVED and REVISED by: Thuy L. Nguyen, MS
TITLE: Chemist
ORG: ERB III/EFED/OPP
TEL: 703/605-0562

SIGNATURE: EFED Share drive Copy

2087116

TEXT SEARCHABLE DOCUMENT 2011

## CONCLUSIONS

### Degradation - Hydrolysis

1. This study is scientifically valid and provides useful information on the hydrolysis of pymetrozine in pH 1, 5, 7, and 9 sterile aqueous buffers at 25°C, 50°C, and 70°C, and on the formation and decline of its degradates.

2. It was noted that data in the submitted studies were reported as distribution of recovered radioactivity from individual analyses, and not as percentages of nominal application rate as specified by the Subdivision N Guidelines for the fulfillment of EPA data requirements. However no data resubmission is needed, and the percentages of the nominal application could be derived from actual concentrations which were reported.

   Overall, the submitted studies can be used to fulfill the EPA data requirements on the hydrolysis of pymetrozine. No additional data is required.

3. Triazine ring-labeled [6-$^{14}$C]pymetrozine, at a nominal concentration of 5 mg/L, was hydrolytically stable in sterile aqueous pH 7 and pH 9 buffer solutions, and hydrolyzed with a calculated half-life of 13.2 days in sterile aqueous pH 5 buffer solution incubated in darkness at 25 °C for up to 32 days. All data were reported as fractions of the total radioactivity recovered from individual analyses unless otherwise noted. At 25 °C, the parent was initially present in the pH 5 buffer solutions at 95.8% (4.92 ppm) of the recovered radioactivity, decreased to 55.1% (2.83 ppm) by 20 days, and was 44.7% (2.30 ppm) at 32 days posttreatment. The major degradate CGA 215525 was present in the pH 5 buffer solution at a maximum of 47.7% at 32 days posttreatment. The minor degradate CGA 249257 was present at a maximum of 2.6% of the recovered radioactivity at 25 days posttreatment. Additional studies conducted at pH 1 (25 °C) and at 50 °C and 70 °C demonstrated that the degradation rate of the test compound increased as the incubation temperature increased. A supplemental study (MRID 44024962) conducted at pH 5 and 25 °C to investigate the pattern of formation and decline of degradates yielded results similar to those of the definitive study, with a registrant-calculated half-life of 12.1 days. In solutions incubated at 25 °C, the parent was initially present at 97.7% (4.31 ppm; single replicate) of the applied radioactivity, decreased to 50.0% (2.23 ppm) by 14 days posttreatment, and was 37.7% (1.65 ppm) at 35 days posttreatment.

## METHODOLOGY

Triazine ring-labeled [6-$^{14}$C]pymetrozine {CGA-215944; 6-methyl-4-(pyridin-3-yl-methyleneamino)-4,5-dihydro-1,2,4-triazin-3(2H)-one; radiochemical purity >96%, specific activity 2.0 Mbq/mg; p. 19}, dissolved in methanol, was added at a nominal concentration of 5 mg/L to a bulk solution of sterile aqueous 0.1 M

TEXT SEARCHABLE DOCUMENT 2011



pH 1 (hydrochloric acid), 0.01 M pH 5 (acetate), 0.01 M pH 7 (phosphate), and 0.01 M pH 9 (borate) buffer solutions (pp. 23-25). Buffer solutions were autoclaved and tested for sterility on agar plates. Treated buffer solutions were transferred to autoclaved glass vials, and the test vials were covered, placed in an incubator or a water bath maintained at 25 °C, 50 °C or 70 °C, and continuously shaken for up to 30 days in darkness. Duplicate samples incubated at 25 °C were collected for analysis at 0, 0.5, 1, 3, 6, 24, and 48 hours posttreatment for the pH 1 samples; at 0, 5, 10, 15, 20, 25, and 30 days posttreatment for the pH 5 samples; and at 0, 5, 9, 15, 20, 25, and 30 days posttreatment for the pH 7 and pH 9 samples (p. 26). Single samples incubated at 50 °C were collected for analysis at 0, 0.5, 1, 2, 3, 6, and 24 hours posttreatment for the pH 1 samples; at 0, 0.25, 1, 2, 4, 7, 10 days for the pH 5 samples; and at 0, 5, 10, 15, 20, 25, and 30 days posttreatment for the pH 7 and 9 samples. Single samples incubated at 70 °C were collected for analysis at 0, 0.5, 1, 2, 3, 6, and 24 hours posttreatment for the pH 1 samples; at 0, 0.5, 1, 3, 6, 24, and 48 hours posttreatment for the pH 5 samples; at 0, 5, 10, 15, 20, 25, and 30 days posttreatment for the pH 7 samples; and at 0, 1, 3, 7, 10, 15, and 20 days posttreatment for the pH 9 samples. Additional sample sets (pH 1 and pH 5 systems only) were prepared as described above and maintained at 25 °C; these sample sets were used to characterize degradation products and to determine additional half-life estimations.

At each sampling interval, duplicate aliquots of the samples were analyzed for total radioactivity by LSC; the limit of detection was twice the background level and the limit of quantitation was three times the background level (p. 27). Samples from the pH 1 and pH 5 systems maintained at 70 °C were adjusted to pH 7 (sodium borate) upon sampling (p. 27; see Comment #7). Samples were analyzed by TLC on Kieselgel 60 $F_{254}$ plates developed in acetone:methanol (4:1, v:v) or butanol:water:acetic acid (4:1:1, v:v:v). Radioactive areas on the plates were quantified using a linear analyzer (p. 28). To characterize radioactive residues, selected samples from the pH 5 system (25 °C) were analyzed by two-dimensional TLC developed using ethyl acetate:methanol:water (12:6:1, v:v:v) and methanol:chloroform:water (12:6:1, v:v:v) or dichloromethane:methanol:acetic acid (9:1:1, v:v:v) and ethyl acetate:ethanol:25% aqueous ammonia solution:water (65:23:4:12, v:v:v:v); radiolabeled compounds were detected with radioimaging scanning. Samples were co-chromatographed with nonradiolabeled reference standards which were located with UV light (wavelength unspecified; p. 29). Samples were further analyzed by reverse-phase HPLC with multiple mobile phase gradients: (Method 1; Nucleosil 120-7 CN column) potassium phosphate:acetonitrile (95:5, v:v; isocratic), (Method 2; Nucleosil 120-5C18 column and Method 3; Spherisorb S5ODS2 column) water:acetonitrile, and (Method 4; Zorbax SB-C18 column) water with 0.05% trifluoroacetic acid:acetonitrile with 0.05% trifluoroacetic acid; radiolabeled compounds were quantified by radioactive flow detection (pp. 30-32). Eluate fractions were collected and analyzed by LSC; column recoveries were generally >95% (p. 32). Samples were co-chromatographed with nonradiolabeled reference standards which were located using UV detection (p. 33). To confirm compound identities, selected samples were also analyzed using GC/MS in the EI mode (p.35).

In a supplemental study (MRID 44024962), triazine ring-labeled [6-$^{14}$C]pymetrozine,

TEXT SEARCHABLE DOCUMENT 2011



dissolved in acetonitrile, was added at a nominal concentration of 4.4 mg/L to autoclaved, silylated glass vials containing sterile aqueous pH 5 (acetate) 0.01 M buffer solution (pp. 15, 18). The buffer solution was sterilized by filtration (0.2 µm). The test vials were sealed, covered with aluminum foil, and maintained at 25 ± 1 °C for up to 35 days in darkness (p. 19). Duplicate samples plus a third sample for sterility confirmation were removed for analysis at 0, 3, 7, 9, 11, 14, 22, 30, and 35 days posttreatment (p. 20). The pH ranged from 4.9 to 5.1 throughout the incubation period (p. 34). At each sampling interval, triplicate aliquots were analyzed for total radioactivity by LSC; the limit of detection ranged from 30 dpm to 68 dpm per sample (p. 28). Samples were analyzed by two-dimensional TLC on silica gel plates developed sequentially with ethyl acetate:methanol:water (75:15:10, v:v:v) and chloroform:methanol:ammonium hydroxide:water (80:30:2:4, v:v:v:v; p. 22). Radioactive areas on the plates were quantified using a radioactive image scanner (p. 23). Samples were co-chromatographed with nonradiolabeled reference standards which were detected by UV light (254 nm; p. 22). Samples were further analyzed by reverse-phase HPLC (Zorbax RX-C8 column) with a mobile phase gradient of 0.05% trifluoroacetic acid in water:0.05% trifluoroacetic acid in methanol (100:0 to 93:7 to 88:12 to 82:18 to 2:98, v:v) using radioactive flow detection (p. 26). Samples were co-chromatographed with nonradiolabeled reference standards which were detected by UV light (260 nm; pp. 26-27). Eluate fractions were collected and analyzed for total radioactivity by LSC. Selected samples (day 7 replicates) were concentrated by solid phase extraction (SPE; Bond Elut column). The column was eluted with hexane followed by acetonitrile and methanol. Selected eluate fractions were collected and analyzed by two-dimensional TLC as previously described and LC/MS (Intersil 5 ODS2 column; pp. 28-29). All samples were analyzed on the day of sampling with the exception of the day 0 sample (replicate 1) which was stored frozen (<5 °C) for 8 days prior to analysis (p. 21).

## DATA SUMMARY

Triazine ring-labeled [6-$^{14}$C]pymetrozine (radiochemical purity >96%), at a nominal concentration of 5 mg/L, was hydrolytically stable in sterile aqueous pH 7 and pH 9 buffer solutions, and hydrolyzed with respective registrant-calculated half-lives of 2.9 hours and 13.2 days in sterile aqueous pH 1 and pH 5 buffer solutions incubated in darkness at 25 °C for up to 32 days (Table 17, p. 58). However, the reported half-lives were questionable since >50% of the radioactivity present at day 0 remained as parent at 3 and 25 days posttreatment for the pH 1 and pH 5 buffer solutions, respectively; the reviewer calculated half-life at pH 1 was 4.5 hours ($r^2$ = 0.99; 0-6 hour data from one data set) based on concentration data in Table 13 (p. 54; see Comment #1). The degradation of the parent in the pH 1 buffer solution was biphasic with an initial rapid degradation phase from 0-6 hours posttreatment (Table 13, p. 54). At pH 5 (25 °C), the reviewer calculated half-life (for the second of two data sets only) was 28.4 days ($r^2$ = 0.98) based on concentration data. Calculated half-lives were also questionable based on those reported in MRID 44024964, in which hydrolysis of the parent was studied using a different radiolabel.

TEXT SEARCHABLE DOCUMENT 2011



The study was also conducted at 50 °C and 70 °C; the data indicated that the degradation rate of the test compound increased as the incubation temperature increased. The parent compound hydrolyzed with respective registrant-calculated half-lives of 0.5 hours, 2.2 days, 79 days ($r^2 = 0.995$), and 44 days ($r^2 = 0.99$) in sterile aqueous pH 1, pH 5, pH 7, and pH 9 buffer solutions incubated in darkness at 50 °C. The parent compound hydrolyzed with respective registrant-calculated half-lives of 0.1 hours, 13 hours ($r^2 = 0.99$), 16 days ($r^2 = 1.0$), and 6.2 days ($r^2 = 0.99$) in sterile aqueous pH 1, pH 5, pH 7, and pH 9 buffer solutions incubated in darkness at 70 °C.

Unless otherwise noted, all data were reported as fractions of the total radioactivity recovered from individual analyses rather than as percentages of the applied or nominal radioactivity (see Comment #2). Only data from the second of two data sets were reported for the pH 1 and pH 5 systems; degradates were not quantified for the first data set at each pH and means could not be accurately determined due to data reporting problems. In the solutions incubated at 25 °C, the parent was initially present in the pH 1 buffer solution at 94.3% (4.84 ppm) of the recovered radioactivity, decreased to 57.5% (2.96 ppm) by 3 hours posttreatment, and was 22.0% (1.10 ppm) at 48 hours posttreatment (bottom of Table 9, p. 50; bottom of Table 13, p. 54). At pH 5, the parent was initially present at 95.8% (4.92 ppm), decreased to 55.1% (2.83 ppm) by 20 days posttreatment, and was 44.7% (2.30 ppm) at 30 days posttreatment. At pH 7 and 9, the parent was hydrolytically stable; the parent compound was present in the pH 7 and pH 9 buffer solutions at 93.9% (4.87 ppm) and 95.4% (5.12 ppm) of the recovered radioactivity, respectively, at 30 days posttreatment (Table 10, p. 51; Table 14, p. 55).

In the pH 5 buffer solution incubated at 25 °C, the major degradate

4-amino-6-methyl-4,5-dihydro-2H-(1,2,4)-triazin-3-one (CGA 215525)

was initially present (day 0) at 0.7% of the recovered radioactivity, increased to 24.2% by 10 days posttreatment, and was a maximum of 47.7% at 32 days posttreatment (Table 19, p. 60). The minor degradate 6-methyl-4,5-dihydro-2H-(1,2,4)-triazin-3-one (CGA 249257) was present at a maximum of 2.6% of the recovered radioactivity at 25 days posttreatment. Three unidentified degradates were each present at ≤1.1% of the recovered radioactivity. Uncharacterized radioactivity was a maximum of 4.0% of the recovered radioactivity at 25 days posttreatment.

In the pH 1 buffer solution incubated at 25 °C, the minor degradate 4-amino-6-methyl-2H-(1,2,4)-triazine-3,5-dione (CGA 294849) was present (means of two replicates) at a maximum of 3.3% of the recovered radioactivity at 6 hours posttreatment (Table 21, p. 62). Radioactivity identified as the combined degradates CGA 249257 and CGA 215525 was present at a maximum of 47.0% of the recovered radioactivity at 24 hours posttreatment. An unidentified compound (Peak HT1) was present at a maximum of 14.4% of the recovered radioactivity at 48 hours posttreatment (Table 21, p. 62; Figure 32, p. 97). A second unidentified compound (Zone A) was present at a maximum of 11.7% of the recovered radioactivity at 24 hours posttreatment.

TEXT SEARCHABLE DOCUMENT 2011



Uncharacterized radioactivity was present at a maximum of 4.9% of the recovered radioactivity at 6 hours posttreatment.

In the solutions incubated at 50 °C, the parent was initially present in the pH 1 buffer solutions at 95.0% (3.6 ppm) of the recovered radioactivity, decreased to 54.0% (2.06 ppm) by 0.5 hours posttreatment, and was 9.8-11.8% (0.37-0.45 ppm) from 2 to 24 hours posttreatment (Table 11, p. 52; Table 15, p. 56). The parent was initially present in the pH 5 buffer solutions at 96.4% (5.02 ppm) of the recovered radioactivity, decreased to 59.2% (3.08 ppm) by 2 days posttreatment, and was 17.1% (0.89 ppm) at 10 days posttreatment. The parent was present in the pH 7 buffer solutions at 96.8% (4.93 ppm) of the recovered radioactivity and decreased to 73.4% by 30 days posttreatment. At pH 9, the parent was initially present at 96.6% (5.04 ppm) of the recovered radioactivity and decreased to 61.9% (3.23 ppm) by 30 days posttreatment.

In the solutions incubated at 70 °C, the parent was initially present in the pH 1 buffer solution at 96.1% (4.98 ppm) of the recovered radioactivity, decreased to 9.9% (0.51 ppm) by 0.5 hours posttreatment, and was 7.3-7.8 (0.38-0.40 ppm) from 1 to 24 hours posttreatment (Table 12, p. 53; Table 16, p. 57). The parent was initially present in the pH 5 buffer solutions at 96.4% (5.0 ppm) of the recovered radioactivity, decreased to 63.8% (3.3 ppm) by 6 hours posttreatment, and was 8.8% (0.46 ppm) at 48 hours posttreatment. The parent was initially present in the pH 7 buffer solution at 96.3% (5.0 ppm) of the recovered radioactivity, decreased to 52.2 (2.7 ppm) by 15 days posttreatment, and was 26.1% (1.4 ppm) at 30 days posttreatment. At pH 9, the parent was initially present at 96.7% (5.2 ppm) of the recovered radioactivity, decreased to 37.6% (2.0 ppm) by 7 days posttreatment, and was 10.0% (0.54 ppm) at 20 days posttreatment.

Based on LSC analysis, material balances (across all temperatures) for the pH 1, 5, 7, and 9 test systems were 97.7-100.0%, 100.3-102.3%, 93.2-101.8%, and 92.7-103.5%, respectively, throughout the incubation periods (Tables 1-4, pp. 42-45); material balances were reported for the second data set only for the pH 1 and pH 5 systems.

In a supplemental study (MRID 44024962), similar results to those of the definitive study were observed in pH 5 (acetate) buffer solution, except that all data were expressed as a percentage of the applied radioactivity rather than as a distribution of the recovered radioactivity. Triazine ring-labeled [6-$^{14}$C]pymetrozine, at a nominal concentration of 5 mg/L (actual concentration 4.4 ppm), hydrolyzed with a registrant-calculated half-life of 12.1 days (based on 0-11 day data; $r^2 = 0.99$; $DT_{50} = 12.3$ days) in sterile aqueous pH 5 buffer solution incubated in darkness at 25 °C for up to 35 days (Figure 18, p. 63). The degradation of the test compound was biphasic with an initial rapid degradation phase occurring from 0 to 11 days posttreatment. In solutions incubated at 25 °C, the parent was initially present at 97.7% (4.31 ppm; single replicate) of the applied radioactivity, decreased to 50.0% (2.23 ppm) by 14 days posttreatment, and was 37.7% (1.65 ppm) at 35 days posttreatment (Tables IV, V, pp. 43, 44). Degradate residue concentrations were reported in parent equivalents. The major degradate

TEXT SEARCHABLE DOCUMENT 2011



4-amino-6-methyl-4,5-dihydro-2H-(1,2,4)-triazin-3-one (CGA 215525)

was present at 14.5% (0.64 ppm) of the applied radioactivity at 3 days posttreatment, increased to 31.6% (1.4 ppm) by 11 days posttreatment, was a maximum of 40.0% (1.8 ppm) at 30 days posttreatment, and was 37.7% (1.7 ppm) at 35 days posttreatment. The minor degradate 6-methyl-4,5-dihydro-2H-(1,2,4)-triazin-3-one (CGA 249257) was present at 0.28% (0.012 ppm; single replicate) of the applied radioactivity at 0 days posttreatment and was a maximum of 1.6% (0.071 ppm) at 35 days posttreatment. Five unidentified degradates were present, each at ≤6.5% (0.29 ppm) of the recovered radioactivity. Unidentified radioactivity was present at a maximum of 6.5% of the recovered radioactivity at 22 days posttreatment. Material balances ranged from 89.2-110.2% (based on single replicates) throughout the incubation period (Table IV, p. 43).

COMMENTS

1. The registrant calculated half-life of 13.2 days (Table 17, p. 58; second data set only) for the parent compound incubated in pH 5 buffer solution at 25 °C was questionable since >50% of the parent was present at 25 days posttreatment. The reviewer-calculated half-life was 28.4 days ($r^2 = 0.98$) based on concentration data in Table 13 (p. 54). The reviewer notes that the half-life of the parent at pH 5 was different from that observed in another study (which utilized a different radiolabel). In MRID 44024964, the registrant-calculated half-life (at pH 5; pyridine label) was 5.0 days and the reviewer-calculated half-life was 14.4 days. Dissimilar half-life results for the parent compound between studies (utilizing different radiolabels) indicates that the reported half-lives may be inaccurate. The reviewer notes that the half-life (at pH 5; triazine label) determined in the supplemental study (MRID 44024962) was 12.1 days.

2. The data (designated as "% test solution radioactivity") in MRID 44024963 were not reported as a percentage of the nominal (or actual) application rate, but appeared to be fractions of the total recovered radioactivity recovered from individual analyses. Confirmation by the registrant is necessary. It may not be necessary that the registrant recalculate and report parent and degradate residues as percentages of the nominal radioactivity, however, as actual concentrations were also reported. The reviewer notes that the percentage data presented in the supplemental study (MRID 44024962) were reported correctly.

3. The reviewer notes that, because initial concentrations of the parent varied slightly between samples in the two data sets for the pH 1 and 5 systems, the percentage remaining as parent compound at each interval for the first set did not necessarily relate to the percentage of parent remaining at each interval for the second set. Therefore, the reviewer did not report means for the two sets, but reported data for the second set only.

4. The aqueous solubility of the parent compound was reported as 270 mg/L at 20 °C (pH

TEXT SEARCHABLE DOCUMENT 2011



unspecified; p. 20, MRID 44024963). The study author did not report the solubility of the parent compound at each pH tested.

5. Minimal microbial growth was observed at 30 days posttreatment for the pH 7 and pH 9 systems incubated at 50 °C, indicating possible contamination (Appendix 3, pp. 113-114, MRID 44024963).

6. Unidentified radioactivity present at >10% of the applied radioactivity was not identified for the pH 1 systems incubated at 25 °C. Two areas designated as "Peak HT1" and "Zone a" were not identified (Table 21, p. 62, MRID 44024963). The study author reported that "Peak HT1" was comprised of several components at >10% of the recovered radioactivity (p. 38; Figure 34, p. 101).

7. A phosphate buffer system was utilized to study the test compound in at pH 7 in water (p. 24, MRID 44024963); it is recommended that borate or acetate buffer solutions be utilized to minimize buffer effects.

8. At the time of sampling, the pH of the pH 1 and pH 5 systems was adjusted to pH 7 (sodium borate). The study author reported that the hydrolysis of the parent compound was expected to be rapid under these lower pH conditions while the test compound would be more hydrolytically stable at pH 7 (p. 27, MRID 44024963).

9. Replicate test samples were not utilized in the study for the systems (all pH systems) incubated at 50 °C and 70 °C (p. 25, MRID 44024963). The use of single test samples is generally not considered good laboratory practice; at a minimum, duplicate test samples should be used.

10. The study author reported that the actual concentration of the test compound utilized in the supplemental study was 4.4 ppm (p. 19; MRID 44024962).

11. The study author reported that the temperature-controlled room used for the incubation period was scheduled for maintenance and that the test samples were moved to a different incubation room for six days because a constant temperature in the original room could not be guaranteed during that time (pp. 32, 87; MRID 44024962).

12. The pH ranges of the pH 1, pH 5, pH 7, and pH 9 systems maintained at 25 °C, 50 °C, and 70 °C were 0.91-1.2, 4.8-5.2, 6.4-7.2, and 8.7-9.3, respectively, throughout the incubation period (Tables 5-8, pp. 46-49, MRID 44024963).