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## Effects of dietary zinc level and ractopamine HCl on pork chop tenderness and shelf-life characteristics

### Authors

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# Effects of Dietary Zinc Level and Ractopamine HCl on Pork Chop Tenderness and Shelf-Life Characteristics<sup>1</sup>

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## Summary

A total of 320 finishing pigs (PIC 327 × 1050; initially 216 lb) were utilized to determine the effects of adding Zn to diets containing ractopamine HCl (RAC) on muscle fiber type distribution, fresh chop color, and cooked meat characteristics. Dietary treatments were fed for approximately 35 d and consisted of: a corn-soybean meal-based negative control (CON); a positive control diet with 10 ppm of RAC (RAC+); and the RAC+ diet plus 75, 150, or 225 ppm added Zn from either ZnO or Availa-Zn. Loins from 80 barrow and 80 gilt carcasses were evaluated. No Zn source effect or Zn source × level interactions were observed during the study ( $P > 0.10$ ). Pigs fed the RAC+ had increased ( $P < 0.02$ ) percentage type IIX and a tendency for increased percentage type IIB muscle fibers. Increasing added Zn decreased (linear,  $P = 0.01$ ) percentage type IIA and tended to increase ( $P = 0.09$ ) IIX muscle fibers. On d 1, 2, 3, 4, and 5 of display, pork chops from pigs fed the RAC+ treatment had greater ( $P < 0.03$ )  $L^*$  values (lighter) compared with the CON. On d 0 and 3 of display, increasing added Zn tended to decrease (quadratic,  $P = 0.10$ )  $L^*$  values and decreased (quadratic,  $P < 0.03$ )  $L^*$  values on d 1, 2, 4, and 5. Pigs fed RAC+ had decreased ( $P < 0.05$ )  $a^*$  values (less red) on d 1 and 4 of display and tended to have decreased ( $P < 0.10$ )  $a^*$  values on d 0 and 2 compared with CON pork chops. RAC+ decreased ( $P < 0.001$ ) metmyoglobin reducing ability (MRA) of pork chops on d 5. Chops from pigs fed added Zn had increased (quadratic,  $P < 0.03$ ) MRA on d 3 and 5 of the display period. There was a trend for increased (linear,  $P = 0.07$ ) cooking loss as added Zn increased in RAC diets. In conclusion, RAC+ diets produced chops that were lighter and less red but maintained a higher percentage of surface oxymyoglobin throughout a 5-d simulated retail display. RAC+ reduced MRA at the end of the display period, but supplementing Zn to RAC diets restored MRA to near CON treatment levels at the end of the display period.

Key words: ractopamine HCl, pork color, pork quality, fiber type, pork chop shelf life

## Introduction

Ractopamine HCl (RAC; Paylean; Elanco Animal Health, Greenfield, IN) is frequently added to finishing pig diets to improve growth performance and carcass leanness. When adding RAC to finishing diets, amino acid concentrations should be increased approximately 30% to maximize growth and carcass lean based on results of

<sup>1</sup> Appreciation is expressed to Elanco Animal Health (Greenfield, IN) for providing partial financial support for this experiment.

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several research trials. Little research has been conducted to determine the effects of additional trace mineral concentrations on the response to RAC, but recent studies have observed that added Zn can increase the response to RAC (Akey, 2011<sup>4</sup>, Patience, 2011<sup>5</sup>).

Although these studies provide initial justification to increase the amount of Zn in diets containing RAC, no research has demonstrated the effects of additional Zn in combination with RAC on fresh meat characteristics, including color stability and cooked meat tenderness. Numerous studies have reported that RAC decreases cooked longissimus muscle (LM) tenderness (Dunshea et al., 2005<sup>6</sup>) and alters LM color (Apple et al., 2008<sup>7</sup>), which are the most important variables consumers use when making a purchasing decision and evaluating their eating experience. Both of these characteristics are influenced by muscle fiber type and the metabolic profile associated with these fibers (Ryu and Kim, 2005<sup>8</sup>; Lee et al., 2010<sup>9</sup>). Because RAC has been previously shown to alter muscle fiber types (Aalhus et al., 1992<sup>10</sup>; Depreux et al., 2002<sup>11</sup>), we speculated that the increased growth and carcasses response experienced by pigs supplemented with RAC and Zn may alter the muscle's fiber type distribution and consequently impact shelf-life and tenderness. Therefore, the objective of this study was to evaluate the effects of adding increasing levels of Zn to RAC containing finishing pig diets on muscle fiber type distribution, fresh chop color, and cooked meat characteristics.

## Procedures

The Kansas State University Institutional Animal Care and Use Committee approved the protocol used in this experiment.

A total of 320 finishing pigs (PIC 327 × 1050) with an average initial BW of 216 lb were housed at the Kansas State University Swine Teaching and Research Center. The finishing barn was an environmentally controlled facility with 5-ft<sup>2</sup> slatted-floor pens. Each pen was equipped with a dry self-feeder and a nipple waterer to provide ad libitum access to feed and water. Two consecutive replications of 160 pigs in the same barn were used. Within each replication, there were 80 pens with 2 pigs per pen. Within each replication, the 80 pens were divided into two 40-pen groups with 24 barrow pens

<sup>4</sup> Akey. 2011. Effects of Zinc Source and Level in Paylean Diets on Pig Performance and Carcass Characteristics. Akey Swine Newsletter.

<sup>5</sup> Patience, J.P. 2011. Impact of Zinc Source and Timing of Implementation on Grow-finish Performance, Carcass Composition, and Locomotion Score. Iowa St. Univ. Anim. Ind. Rep.

<sup>6</sup> Dunshea, F.R., D.N. D'Souza, D.W. Pethick, G.S. Harper, and R.D. Warner. 2005. Review: Effects of dietary factors and other metabolic modifiers on quality and nutritional value of meat. Meat Sci. 71:8–38.

<sup>7</sup> Apple, J.K., C.V. Maxwell, B.R. Kutz, L.K. Rakes, J.T. Sawyer, Z.B. Johnson, T.A. Armstrong, S.N. Carr, and P.D. Matzat. 2008. Interactive effect of ractopamine and dietary fat source on pork quality characteristics of fresh pork chops during simulated retail display. J. Anim. Sci. 86:2711–2722.

<sup>8</sup> Ryu, Y.C., and B.C. Kim. 2005. The relationship between muscle fiber characteristics, postmortem metabolic rate, and meat quality of *Longissimus dorsi* muscle. Meat Sci. 71:351–357.

<sup>9</sup> Lee, S.H., S.T. Joo, and Y.C. Ryu. 2010. Skeletal muscle fiber type and myofibrillar proteins in relation to meat quality. Meat Sci. 86:166–170.

<sup>10</sup> Aalhus, J.L., A.L. Schaefer, A.C. Murray, and D.M. Jones. 1992. The effect of ractopamine on myofibre distribution and morphology and their relation to meat quality in swine. Meat Sci. 31:397–409.

<sup>11</sup> Depreux, F.F.S., A.L. Grant, D.B. Anderson, and D.E. Gerrard. 2002. Paylean alters myosin heavy chain isoform content in pig muscle. J. Anim. Sci. 80:1888–1894.

and 16 gilt pens or 16 barrow pens and 24 gilt pens per group, resulting in 4 groups of 40 pens each.

Pens of pigs were allotted to 1 of 8 dietary treatments, with 2 pigs per pen and a total of 20 pens per treatment. Dietary treatments consisted of: a corn-soybean meal–based negative control diet formulated to 0.66% standardized ileal digestible (SID) lysine (CON); a positive control diet formulated to contain 0.92% SID lysine and 10 ppm of RAC (RAC+); the RAC+ diet plus 75, 150, or 225 ppm added Zn from ZnO; and the RAC+ diet plus 75, 150, or 225 ppm added Zn from Availa-Zn (Zinpro, Eden Prairie, MN; Table 1). All diets contained a trace mineral premix that contributed 55 ppm Zn from ZnSO<sub>4</sub>. Experimental diets were fed in meal form, and ZnO or Availa-Zn was added to the RAC diet at the expense of corn. Diets were fed for the last 41 d before slaughter for group 1 and the last 35 days for groups 2, 3, and 4. Treatment effects on growth and carcass composition are reported in another report (see “Effects of Added Zn in Diets with ractopamine HCl on Growth Performance, Carcass Characteristics, and Zn Concentrations in Plasma, Loin, and Liver of Finishing Pigs,” p. 132).

### ***Harvest and sample collection***

At the completion of the feeding period, 1 pig was randomly selected from each pen and transported to the Kansas State University Meats Laboratory for harvest under federal inspection. After a 24-h post-slaughter chilling period, a 12-in. portion of the *longissimus lumborum* muscle (beginning at the 10th rib) was collected from the left side for immunohistochemistry and fresh pork quality analysis. Additionally, 24-h pH was measured using a Hanna HI 99163 meat pH probe (Hanna Instruments, Smithfield, RI) inserted into the 11th–12th-rib interface, and a 1-in.-thick chop was collected from this location to be used for immunohistochemical analysis. The remainder of the muscle sample was vacuum-packaged and stored at 36°F for 13 d postmortem.

### ***Immunohistochemistry***

A 0.39-in.<sup>2</sup> portion of muscle was collected from the geometric center of each chop designated for immunohistochemistry. After collection, the muscle was embedded in Optimal Cutting Temperature (OCT) tissue embedding media (Fisher Scientific, Pittsburgh, PA), frozen by submersion in supercooled isopentane, and stored at -112°F until analysis. For each sample, two 10-μm cryosections were collected on frost-resistant slides (Fisher Scientific), and the methods of Gonzalez et al. (2008<sup>12</sup>) were followed for immunodetection with modifications. Non-specific antigen-binding sites were inhibited by incubating cryosections in 5% horse serum and 0.2% TritonX-100 in phosphate-buffered saline (PBS) for 30 min. All sections were incubated with the following primary antibodies in blocking solution for 60 min: 1:50 α-dystrophin (Thermo Scientific, Waltham, MA); 1:10 supernatant myosin heavy-chain, slow IgG2b (BA-D5, Developmental Studies Hybridoma Bank, University of Iowa, Iowa City, IA); 1:10 supernatant myosin heavy-chain type 2A, IgG1 (SC-71, Developmental Studies Hybridoma Bank); and 1:10 supernatant myosin heavy-chain type 2B, IgM (BF-F3, Developmental Studies Hybridoma Bank, University of Iowa, Iowa City, IA). After incubation, sections were washed with PBS 3 times for 5 min, followed by incuba-

<sup>12</sup> Gonzalez, J.M., S.E. Johnson, T.A. Thrift, J.D. Savell, S.E. Ouellette, and D.D. Johnson. 2009. Effect of ractopamine-hydrochloride on the fiber type distribution and shelf-life of six muscles of steers. *J. Anim. Sci.* 87:1764–1771.

tion in the following secondary antibodies (1:1000) in blocking solution for 30 min: Alexa-Fluor 488 goat anti-mouse IgM for BF-F3 (Invitrogen, San Diego, CA); Alexa-Fluor 594 goat anti-mouse IgG1 for SC-71 (Invitrogen); Alexa-Fluor 633 goat anti-mouse IgG2b for BA-D5 (Invitrogen); and Alexa-Fluor 594 goat anti-rabbit H&L for  $\alpha$ -dystrophin (Invitrogen). In addition, 1:1000 Hoechst Dye 33342 (Invitrogen) was utilized to identify all fiber-associated nuclei. Finally, sections were washed for three 5-min periods in PBS, then covered with 5  $\mu$ L of 9:1 glycerol in PBS, then coverslipped for imaging.

Cryosections were imaged using a Nikon Eclipse TI-U inverted microscope with 10 $\times$  working distance magnification (Nikon Instruments Inc., Melville, NY). Four representative photomicrographs per section were captured using a Nikon DS-QiMc digital camera (Nikon Instruments Inc.) calibrated to the 10 $\times$  objective. For myosin heavy-chain fiber-type data collection, a minimum of 2 photomicrographs per section (minimum 500 fibers per animal) were analyzed for isoform distribution with NIS-Elements Imaging Software (Basic Research, 3.3; Nikon Instruments Inc.). Fibers that were positive for the BA-D5 antibody were counted as type I fibers. Fibers strongly stained only for SC-71 or BF-F3 were labeled as type IIA and type IIB fibers, respectively. Fibers stained weakly for both SC-71 and BF-F3 were labeled as type IIX fibers.

### ***Chop cutting and simulated retail display***

At the conclusion of the 13-d aging period, muscles were removed from the package, oriented in the same direction, and cut into five 1-in.-thick chops. The first 4 chops closest to the 10th rib were utilized for simulated retail display. Of these chops, the first 3 were used for d 0, 1, and 3 metmyoglobin reducing ability analysis, and the fourth chop was used for 5-d chop surface color attributes including the collection of  $L^*$ ,  $a^*$ , and  $b^*$  values and spectral data for the calculation of surface myoglobin redox forms. The fourth chop was also used for d-5 metmyoglobin reducing ability analysis. The last chop was immediately analyzed for mechanical tenderness by Warner-Bratzler shear force.

All chops allocated to simulate retail display were placed on white 1S Styrofoam trays (Genpack, Glens Falls, NY) with a Dri-Loc 50 (Cryovac Sealed Air Corporation, Duncun, SC) absorbent pad and overwrapped with PVC film (MAPAC M [1,450 cc/23.6 in.<sup>2</sup>/24 h, 72 gauge], Bordon Packaging and Industrial Products, North Andover, MA). Chops were placed in coffin-style retail cases (Model DMF 8; Tyler Refrigeration Corporation, Niles, MI) at  $37.4 \pm 3.6^\circ\text{F}$ . Cases were constantly illuminated with fluorescent lights (32 W Del-Warm White 3000° K; Philips Lighting Company, Somerset, NJ) that emitted a 24-h case average intensity of  $2,143 \pm 113$  lx. Every 12 h, chops were rotated from left to right and front to back in the cases to account for variation in temperature and light intensity. Absolute CIE  $L^*$ ,  $a^*$ , and  $b^*$ , and spectral reflectance (400 to 700 nm) readings were taken at 3 locations on each 5-d retail display chop using a Hunter Lab Miniscan EZ spectrophotometer (Illuminant A, 1 in diameter aperture, 10° observer; Hunter Associates Laboratory, Reston, VA). Absolute and spectral values from the 3 scans were averaged and reflectance at 473, 525, 572, and 700 nm



were used to calculate surface percentages of metmyoglobin and oxymyoglobin using the equations published in the AMSA Color Guidelines (AMSA, 2012<sup>13</sup>).

### ***Metmyoglobin reducing ability***

The procedures of Gonzalez et al. (2009<sup>12</sup>) were followed for metmyoglobin reducing ability (MRA) with modifications. On the day of analysis, chops were pulled from the retail display case and cut into 2 × 2-in. portions that were indicative of the discoloration pattern for the entire chop. Each section was placed in a 400-mL beaker and oxidized in 100 mL of 0.3% sodium nitrite at 77 ± 3.6 °F for 20 min. After the samples were blotted of excess solution, they were vacuum-packaged in 10 × 12 in Prime Source Vacuum Pouches (3-mil standard barrier, Bunzl Processor Division, Koch Supplies, Kansas City, MO) that possess an oxygen transmission rate of 4.5 cc/1002/24 h/73 °F/65% relative humidity. Reflectance measurements (400 to 700 nm) were collected initially after vacuum-packaging and every 30 min for 2 h using a Hunter Lab Miniscan EZ spectrophotometer (Illuminant A, 1 in diameter aperture, 10° observer; Hunter Associates Laboratory). Average reflectance values at 525, 572, and 700 nm were used to calculate metmyoglobin percentage at all time points using the equations published in the Meat Color Guidelines, AMSA (2012<sup>13</sup>). Metmyoglobin reducing ability was calculated as: (observed decrease in metmyoglobin concentration ÷ initial metmyoglobin concentration) × 100.

### ***Warner-Bratzler shear force analysis and cooking loss***

The AMSA (1995<sup>14</sup>) guidelines for instrumental cooked meat tenderness were followed for shear force analysis. Fresh-cut chops were weighed and a thermocouple wire (30-gauge and constantan, Omega Engineering, Stamford, CT) was inserted into the geometric center of each chop for internal temperature monitoring using a Doric Minitrend 205 monitor (VAS Engineering, San Francisco, CA). Chops were cooked on electric, open-hearth Farberware grills (Model 450-A; Yonkers, NY) to an internal temperature of 95°F, then flipped and cooked to a final internal temperature of 160°F. After cooked chops were chilled overnight at 45 ± 1.8°F, six 0.5-in.-diameter cores were extracted from each chop parallel to the muscle fiber orientation. Each core was sheared once through the center of the core perpendicular to the muscle fiber orientation with an Instron Model 5569 Testing Machine (Instron, Canton, MA) with a Warner-Bratzler shear head attached (220.5-lb compression load cell, crosshead speed of 9.8 in./min). Cooking loss was determined by measuring the difference in chop weight before and after cooking and dividing by precooked chop weight.

### ***Statistics***

All data were analyzed as a generalized randomized complete block design using the MIXED procedure of SAS (SAS Institute, Inc., Cary, NC) with pen as the experimental unit and animal/chop as the observational unit. For cooked meat analysis and fiber isoform distribution, dietary treatment served as the fixed effect, and gender within group within barn was included as random effects. Contrast statements consisted of: (1) negative control vs. positive control RAC diet, (2) interaction between increasing Zn and Zn source, (3) increasing Zn linear and quadratic polynomials, and (4) added

<sup>13</sup> AMSA (American Meat Science Association). 2012. Meat color measurement guidelines. Am. Meat Sci. Assoc., Champaign, IL.

<sup>14</sup> AMSA (American Meat Science Association). 1995. Research Guidelines for Cookery, Sensory Evaluation, and Instrumental Tenderness Measurements of Fresh Meat. Am. Meat Sci. Assoc., Chicago, IL.

Zn from ZnO vs. Availa-Zn. For shelf-life analysis, the statistical structure was the same, except day of display and the day  $\times$  treatment interaction served as fixed effects in addition to dietary treatment. Day of display  $\times$  dietary treatment interaction was evaluated by the following contrast: (1) interaction between negative control vs. positive control RAC diet and day of display, (2) interaction between increasing Zn and day of display, (3) interaction between Zn source and day of display, and (4) interaction between increasing Zn, Zn source, and day of display. Day of display also served as the repeated measure with chop as the subject. Statistical significance was determined at  $P < 0.05$ , and  $P > 0.05$  and  $P < 0.10$  were termed trends or tendencies.

## Results and Discussion

For all dependent variables observed in the study, there was no evidence of a Zn source effect or an interaction between Zn source and Zn level ( $P > 0.10$ ). Therefore, all data presented will combine the Zn treatment groups by the level of supplementation, making three pooled Zn treatments: 75 (75Zn), 150 (150Zn), and 225 ppm (225Zn) Zn.

Using immunohistochemical techniques, the effects of dietary treatments on myosin heavy-chain isoform distribution were evaluated (Figure 1). This analysis evaluated the four different muscle fiber types: slow-oxidative (type I), fast oxido-glycolytic (type IIA), and fast glycolytic IIX or IIB. Our method of muscle fiber type determination is in agreement with Lefaucheur et al. (2002), who used the same set of antibodies in the LM of the pig. Similar to our findings, these authors reported that SC-71 recognized both type IIA and IIX fibers, with the IIA fibers staining more intensely than the IIX fibers. In addition, our fiber isoform distribution pattern was similar to the distribution reported by the authors, with a high percentage of type IIB fibers (46 to 50%), a moderate percentage of type IIX fibers (25 to 32%), and low percentages of type I (8%) and IIA (14 to 11%) fibers.

Our data indicate that feeding RAC+ or RAC plus added Zn diets did not affect ( $P > 0.62$ ) the percentage of type I muscle fibers. No evidence ( $P = 0.16$ ) was observed for a difference in the percentage of type IIA fibers when comparing muscles from pigs fed CON and RAC+ diets; however, the percentage of type IIA fibers decreased (linear,  $P = 0.01$ ) as dietary Zn increased. Loin muscle samples from pigs fed the RAC+ treatment had a decreased ( $P = 0.02$ ) percentage of type IIX muscle fibers compared with CON; however, adding Zn to the RAC diet tended to increase (linear,  $P = 0.09$ ) the percentage of type IIX fibers. For type IIB fibers, pigs fed the RAC+ treatment had a tendency to possess more ( $P = 0.10$ ) fibers than CON pigs. Finally, no evidence was found ( $P > 0.70$ ) that added Zn affected percentage of type IIB muscle fibers in the loin.

Pork chops from all treatment groups were displayed under simulated retail conditions for 5 d, and daily objective measures of pork color were collected. For  $L^*$ ,  $a^*$ , and  $b^*$  values (Figure 2), only  $a^*$  and  $b^*$  values were affected by day of display (quadratic,  $P < 0.05$ ). Pork chops of pigs from the RAC+ treatment did not differ ( $P > 0.13$ ) in  $L^*$  values compared with those from pigs fed the CON on d 0 of the display period. On d 1, 2, 3, 4, and 5, pork chops from pigs fed the RAC+ treatment had greater  $L^*$  values than those from pigs fed the CON treatment ( $P < 0.03$ ). On d 0 and 3 of display, increasing the Zn content of the diet resulted in a trend for lower  $L^*$  values (quadratic,



$P = 0.10$ ). On d 1, 2, 4, and 5 of the display period, increasing Zn reduced the  $L^*$  values (quadratic,  $P < 0.03$ ). Of the Zn treatment groups, pork chops from pigs fed 150Zn possessed the lowest  $L^*$  values over the entire display period. Pork chops from pigs fed RAC+ had lower  $a^*$  values compared with those from the CON fed pigs on d 1 and 4 of display ( $P < 0.05$ ); however, RAC+ produced chops with  $a^*$  values that tended to be lower on d 0 and 2 of display ( $P < 0.10$ ). On d 4 of display,  $a^*$  values increased (quadratic,  $P = 0.04$ ) as Zn was added to the diet, with chops from the 150Zn treatment possessing the greatest  $a^*$  value. For all other display days, there was a lack of evidence ( $P > 0.12$ ) that increasing dietary Zn influenced  $a^*$  values. On d 0 of display, there was no evidence ( $P > 0.28$ ) for differences in the  $b^*$  values of pork chops from RAC+ fed pigs compared with CON-fed pigs; however, for the remainder of the display period, chops from RAC+-fed pigs possessed lower  $b^*$  values than CON-fed pigs ( $P < 0.04$ ). On d 1 of display, adding dietary Zn increased (quadratic,  $P = 0.03$ )  $b^*$  values, and on d 2 and 4, added Zn tended to increase  $b^*$  values ( $P < 0.09$ ). Chops from pigs fed RAC+ from our study appeared less red and less blue than those from CON-fed pigs throughout most of the display period, and chops from pigs fed RAC+ were lighter than the CON chops on d 1–5 of retail display. This result can be explained by a trend toward increased type IIB fibers in chops from pigs fed RAC+ compared with those fed the CON diet. Interesting quadratic Zn effects were detected on d 3 for  $a^*$  values, d 1–5 for  $L^*$  values, and d 1, 2, and 4 for  $b^*$  values. These results indicate that when Zn was added to the RAC diet, values for each parameter shifted away from RAC+ values and toward the CON values. In addition, because Zn supplementation decreased the amount of type IIA fibers and these fibers contain more myoglobin than IIX and IIB fibers, we believe this observed added Zn effect was independent of muscle fiber shifts.

We further explored the effects of RAC on chop shelf-life by utilizing the equations of Krzywicki (1979<sup>15</sup>) to measure the percentage formation of oxymyoglobin and metmyoglobin on the surface of chops and their associated MRA during display. The objective measures of chop surface oxymyoglobin and metmyoglobin percentages indicated a day effect on both redox forms ( $P < 0.001$ ; Figure 3). Oxymyoglobin surface percentages increased (quadratic,  $P < 0.001$ ) from d 0 to d 1, but the surface percentage of oxymyoglobin decreased thereafter. On d 0 of the display period, we observed no evidence ( $P = 0.63$ ) of differences in amount of oxymyoglobin formed on the surface of chops from pigs fed CON or RAC+; however, on the same day of display, as dietary Zn increased from 0 to 225 ppm in RAC diets, formation of pork chop surface oxymyoglobin percentage decreased (linear,  $P < 0.01$ ). For the remaining days of display, RAC+-treated pigs had chops with a tendency toward increased oxymyoglobin percentage compared with chops from CON-fed pigs ( $P < 0.08$ ). As the day of display increased, the surface percentage of metmyoglobin increased (quadratic,  $P < 0.001$ ), but no evidence was found that including dietary RAC or increasing Zn affected chop surface metmyoglobin accumulation ( $P > 0.10$ ). Full bloom for all treatments, as indicated by the peak oxymyoglobin formation, was reached on d 1 of display, which follows the same pattern as  $a^*$  values. At this time point, RAC+ chops tended to possess more oxymyoglobin on their surface, and this finding was maintained on all days of display except d 3. We attributed these findings to RAC+ chops reaching a

<sup>15</sup> Krzywicki, K. 1979. Assessment of relative content of myoglobin, oxymyoglobin and metmyoglobin at the surface of beef. Meat Sci. 3:1–10.

higher maximum bloom on d 1 and all of the dietary treatments decreasing in surface oxymyoglobin percentage at the same relative rate throughout the display period (Table 2). Our finding that RAC+ chops contain more surface oxymyoglobin than CON chops seems counterintuitive considering RAC+ chops were lighter and less red according to  $L^*$  and  $a^*$  values. We attribute the increased surface oxymyoglobin to RAC+ pigs tending to possess more type IIB fibers. Because of this shift, we are certain that the amount of myoglobin and oxygen-scavenging mitochondria present in the muscle was reduced (Aberle et al., 2003<sup>16</sup>). When muscle possesses a copious amount of mitochondria, myoglobin must compete with these organelles for oxygen, which results in less oxymyoglobin formation (Klont, 1998<sup>17</sup>); therefore, because the RAC+ chops possessed a fiber type distribution that favored the presence of fewer mitochondria, more oxygen was available for consumption by the myoglobin in the muscle, which increased the formation of oxymyoglobin.

As expected, all chops exhibited a reduction ( $P < 0.001$ ) in MRA as the day of display increased (Figure 4). At the beginning of the display period, chop reducing ability ranged from 52% to 56%. No evidence of an RAC+ effect was observed on the reducing ability on this day or d 1 and 3 of the display period ( $P > 0.10$ ). By d 5 of display, chop reducing abilities ranged from 31% to 42%. Inclusion of RAC in the diet reduced ( $P < 0.001$ ) metmyoglobin reducing ability compared with the chops from CON-fed pigs. Although there was no evidence of a Zn effect on d 0 and 1 ( $P > 0.10$ ), as dietary Zn increased, reducing ability increased on d 3 (quadratic,  $P = 0.03$ ) and d 5 (quadratic,  $P = 0.02$ ) of the display period. Seventy-five ppm of added Zn was sufficient to maximize the reducing ability of RAC-treated pork chops on d 3 of display, whereas 150 ppm of added Zn resulted in the greatest reducing ability on d 5. The RAC effect on MRA was not detected until d 5 of the display period, when RAC+ chops possessed 11.6% less MRA than CON chops. At this time point, a quadratic Zn effect was observed in which adding 150 ppm of Zn increased MRA by 9.2% over RAC+ chops. This same effect was seen at d 3, where adding 75 ppm of Zn to the diet increased MRA by 6.3% over RAC+ chops. The fact that RAC stimulated a shift toward type IIB fibers and this did not affect MRA until d 5 of display could indicate that the fiber shift influence on MRA does not become important until later in the display period, when this attribute is the most important to a retailer. In addition, the Zn75 treatment group had the highest MRA and type IIA fiber percentage of the Zn treatments. Because type IIA fibers are more glycolytic than IIX or IIB fibers, more mitochondria and NADH could be available, causing this treatment to have a greater MRA. Although the difference between RAC+ and CON MRA detected at d 5 did not translate to increases in chop surface metmyoglobin formation, extending the display period could demonstrate that the RAC-induced reduction in MRA may result in greater metmyoglobin formation on the surface of these chops. Furthermore, if extending the display period proves that RAC reduces color stability during extended display, Zn supplementation can serve as a countermeasure to these effects, as indicated by the ability of the Zn treatments to restore MRA and  $a^*$  values close to control values.

<sup>16</sup> Aberle, E.D., J.C. Forrest, D.E. Gerrard, and E.W. Mills. 2001. Principles of Meat Science. 4<sup>th</sup> ed. Kendall/Hunt Publishing Co. Dubuque, IA.

<sup>17</sup> Klont, R.E., L. Brocks and G. Eikelenboom. 1998. Muscle fibre type and meat quality. Meat Sci. 49:S219–S229.

We found no evidence ( $P > 0.30$ ) that RAC+ elicited different pH, cooking loss, or shear force values compared with the CON treatment, but we observed a trend toward increased (linear,  $P = 0.07$ ) cooking loss as dietary Zn increased in RAC containing diets (Table 3).

In summary, feeding pigs 10 ppm of RAC decreased the amount of type IIX fibers while tending to increase the percentage of type IIB fibers in the *Longissimus lumborum*. Supplementing the RAC diets with dietary Zn above that contained in the trace mineral premix decreased the percentage of type IIA fibers and tended to increase the percentage of type IIX fibers, which affected meat color characteristics. Pigs fed RAC produced chops that were lighter and less red, but maintained a higher percentage of surface oxymyoglobin throughout a 5-d simulated retail display. Although RAC improved these shelf-life characteristics, it reduced MRA at the end of the display period. Supplementing Zn to RAC diets restored MRA to near-CON treatment levels at the end of the display period, which is most important to retailers. Zinc supplementation tended to increase chop cook loss, which may affect sensory attributes of the chops and should be explored further.

**Table 1. Diet composition (as-fed basis)<sup>1,2</sup>**

Item	Control	RAC
Ingredient, %		
Corn	83.06	74.24
Soybean meal, (46.5% CP)	15.22	23.97
Monocalcium P, (21% P)	0.25	0.20
Limestone	0.75	0.78
Salt	0.35	0.35
Vitamin premix	0.075	0.075
Trace mineral premix <sup>3</sup>	0.075	0.075
L-lysine HCl	0.15	0.15
DL-methionine	---	0.015
L-threonine	---	0.025
Phytase <sup>4</sup>	0.075	0.075
Ractopamine HCl <sup>5</sup>	---	0.05
Total	100	100
Calculated analysis, %		
Standardized ileal digestible (SID) amino acids, %		
Lysine	0.70	0.92
Isoleucine:lysine	71	70
Leucine:lysine	179	158
Methionine:lysine	31	30
Met & Cys:lysine	65	60
Threonine:lysine	63	64
Tryptophan:lysine	19	19
Valine:lysine	84	79
Total lysine, %	0.79	1.03
CP, %	14.3	17.6
ME, Kcal/lb <sup>6</sup>	1,525	1,523
NE, Kcal/lb <sup>7</sup>	1,044	1,029
SID lysine: ME, g/Mcal	2.08	2.74
Ca, %	0.41	0.44
P, %	0.39	0.42
Available P, %	0.21	0.21

<sup>1</sup>Diets were fed in meal form during the experiment.

<sup>2</sup>Dietary treatments were obtained by replacing corn in the ractopamine HCl diet to achieve 75, 150, and 225 ppm added Zn from ZnO (Zinc Nacional S.A., Monterrey, Mexico) or Availa-Zn (Zinpro, Eden Prairie, MN).

<sup>3</sup>Trace mineral premix provided 55 ppm Zn from ZnSO<sub>4</sub>.

<sup>4</sup>Phyzyme 600 (Danisco Animal Nutrition, St. Louis, MO) provided 204 phytase units (FTU)/lb, with a release of 0.10% available P.

<sup>5</sup>Provided 9 g/lb (10 ppm) of ractopamine HCl (Paylean; Elanco Animal Health, Greenfield, IN).

<sup>6</sup>ME values for ingredients were derived from NRC (1998).

<sup>7</sup>NE values for all ingredients were derived from INRA (2004).

**Table 2. LSMEANS of the daily reduction in surface oxymyoglobin percentage from d 1 of display of pork *Longissimus lumborum* chops from pigs supplemented ractopamine HCl and 3 levels of dietary zinc**

Day of display	Control	RAC+ <sup>1</sup>	Zinc, ppm <sup>1,2</sup>				SEM	RAC	P-value	
			75	150	225				Zn linear	Zn quadratic
2	-7.66	-7.19	-6.88	-6.52	-7.39	1.18		0.72	0.95	0.41
3	-11.63	-10.92	-9.63	-9.53	-10.77	1.18		0.58	0.87	0.08
4	-15.47	-14.90	-13.76	-14.06	-15.11	1.18		0.66	0.80	0.13
5	-18.01	-17.39	-16.73	-16.36	-17.50	1.18		0.63	0.99	0.21

<sup>1</sup>Diets contained 10 ppm of ractopamine-HCl (Paylean; Elanco Animal Health, Greenfield, IN).

<sup>2</sup>Dietary treatments were obtained by replacing corn in the ractopamine HCl diets to achieve 75, 150, and 225 ppm added Zn from ZnO (Zinc Nacional S.A., Monterrey, Mexico) or Availa-Zn (Zinpro, Eden Prairie, MN). Because no Zn × source interaction was observed, means represent the pooled results of both Zn sources.

**Table 3. LSMEANS of pork *Longissimus lumborum* chop cooked meat characteristics from pigs supplemented ractopamine HCl and 3 levels of dietary zinc**

Item	Control	RAC+ <sup>1</sup>	Zinc, ppm <sup>1,2</sup>				SEM	RAC	P-value	
			75	150	225				Zn linear	Zn quadratic
Cooking loss, %	24.74	23.54	25.06	24.60	25.63	0.98		0.30	0.07	0.70
Shear force, lb	7.85	7.83	8.29	8.00	8.22	0.31		0.97	0.44	0.59
pH <sup>3</sup>	5.44	5.43	5.44	5.46	5.44	0.02		0.89	0.67	0.43

<sup>1</sup>Diets contained 10 ppm ractopamine HCl (Paylean; Elanco Animal Health, Greenfield, IN).

<sup>2</sup>Dietary treatments were obtained by replacing corn in the ractopamine HCl diets to achieve 75, 150, and 225 ppm added Zn from ZnO (Zinc Nacional S.A., Monterrey, Mexico) or Availa-Zn (Zinpro, Eden Prairie, MN). Because no Zn × source interaction was observed, means represent the pooled results of both Zn sources.

<sup>3</sup>pH collected at 24-h postmortem.



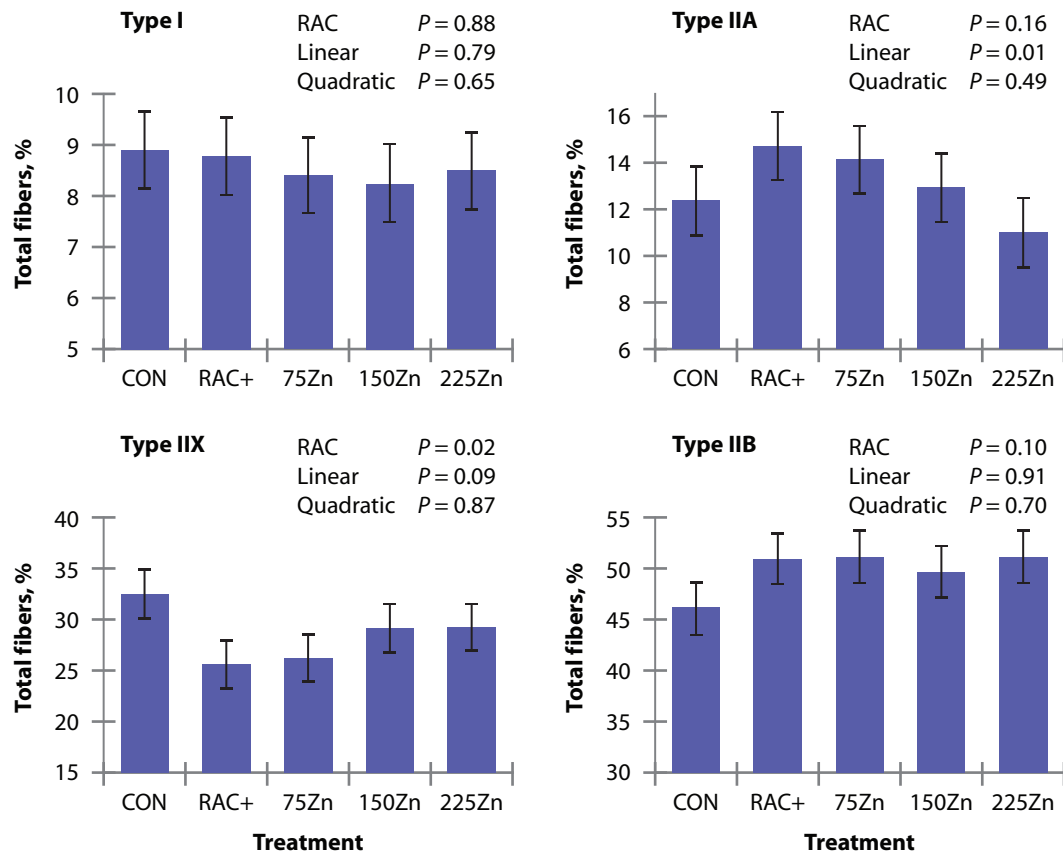


Figure 1. Tenth-rib *Longissimus lumborum* myosin heavy-chain isoform distribution of pigs fed a basal diet containing 0 ppm ractopamine HCl (CON), pigs supplemented 10 ppm ractopamine HCl (RAC+), and pigs supplemented 10 ppm ractopamine HCl and 75 ppm (75Zn), 150 ppm (150Zn), or 225 ppm (225Zn) of zinc.

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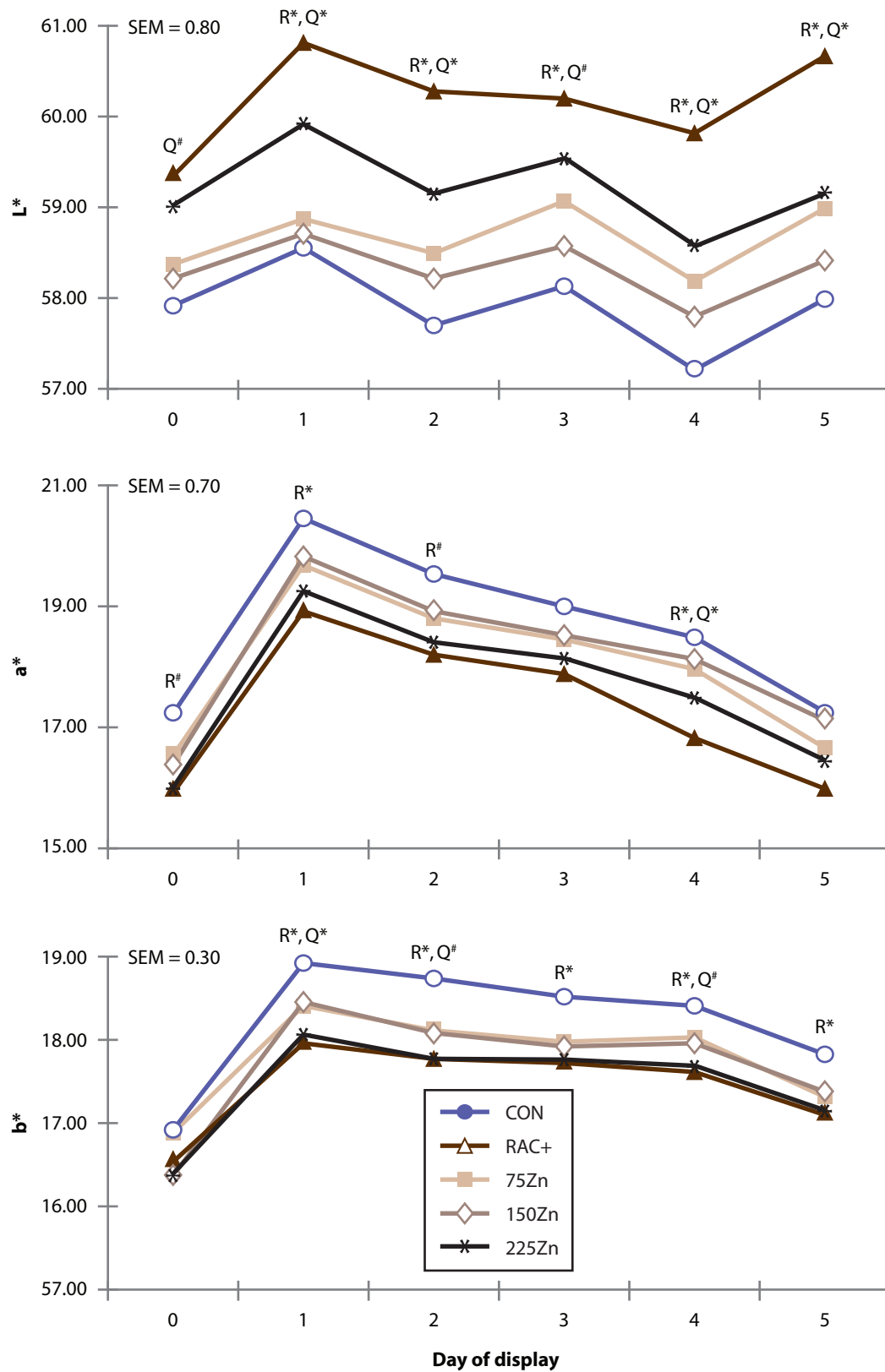


Figure 2. Surface L\*, a\*, and b\* values of loin chops from pigs fed a basal diet containing 0 ppm ractopamine HCl (CON), pigs supplemented 10 ppm ractopamine HCl (RAC+), and pigs supplemented 10 ppm ractopamine HCl and 75 ppm (75Zn), 150 ppm (150Zn), or 225 ppm (225Zn) of zinc. L\* = lightness (0 = black; 100 = white), a\* = redness (-60 = green; 60 = red), and b\* = blueness (-60 = blue; 60 = yellow). R designates a ractopamine-HCl effect and Q designates a quadratic zinc effect. The superscript \* indicates a significant effect ( $P \leq 0.05$ ), and the superscript # indicates marginal significance ( $P \leq 0.10$ ).

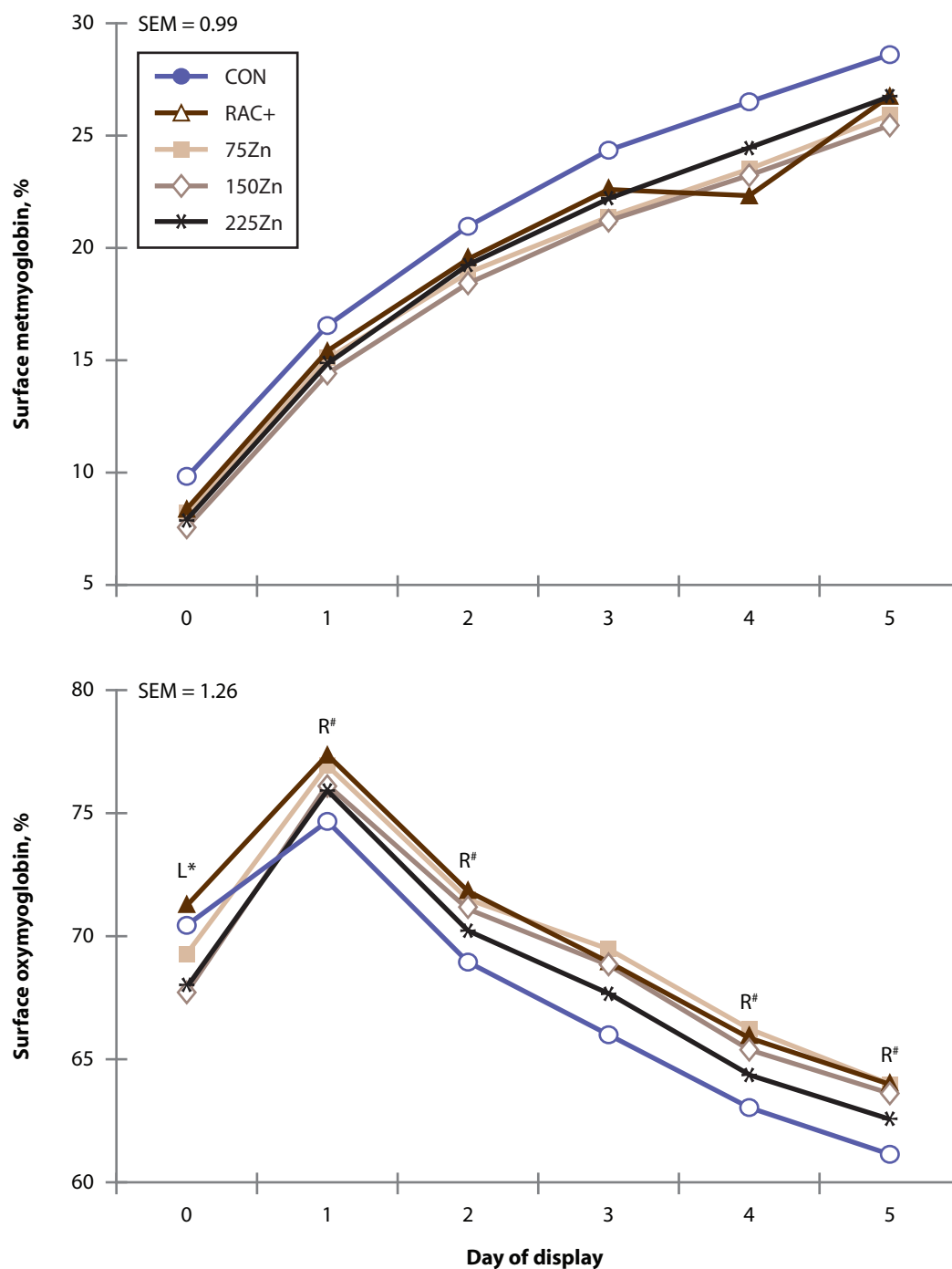


Figure 3. Surface oxymyoglobin and metmyoglobin percentages of loin chops from pigs fed a basal diet containing 0 ppm ractopamine HCl (CON), pigs supplemented 10 ppm ractopamine HCl (RAC+), and pigs supplemented 10 ppm ractopamine HCl and 75 ppm (75Zn), 150 ppm (150Zn), or 225 ppm (225Zn) of zinc. R designates a ractopamine HCl effect and L designates a linear zinc effect. The superscript \* indicates a significant effect ( $P \leq 0.05$ ), and the superscript # indicates marginal significance ( $P \leq 0.10$ ).

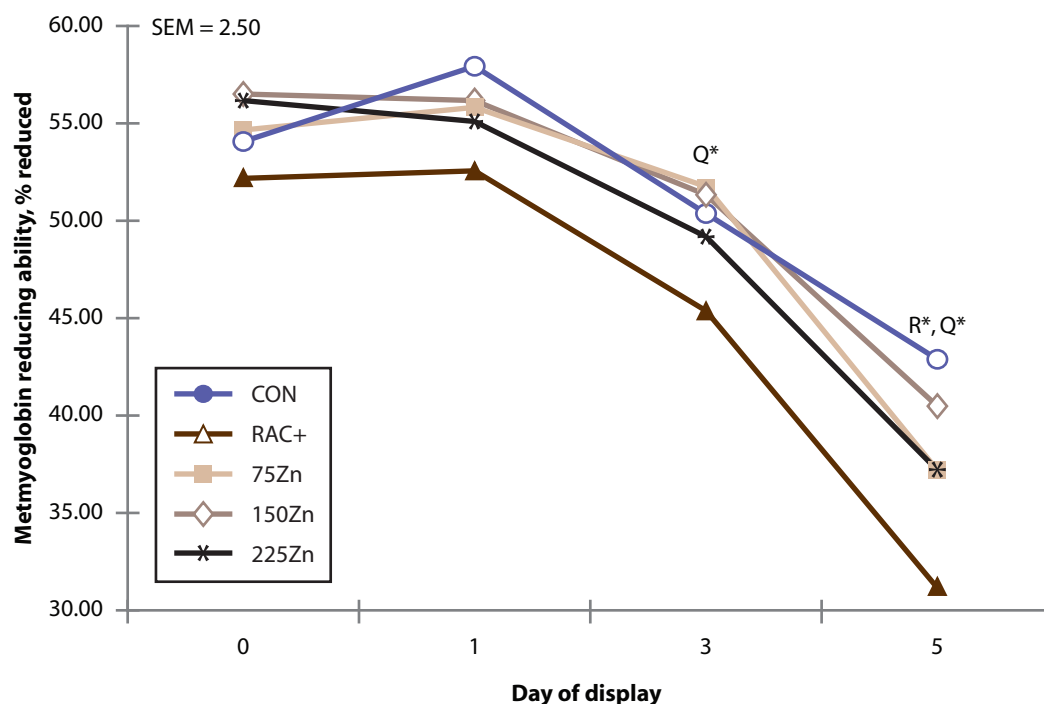


Figure 4. Metmyoglobin reducing ability of loin chops from pigs fed a basal diet containing 0 ppm ractopamine HCl (CON), pigs supplemented 10 ppm ractopamine HCl (RAC+), and pigs supplemented 10 ppm ractopamine HCl and 75 ppm (75Zn), 150 ppm (150Zn), or 225 ppm (225Zn) of zinc. R designates a ractopamine HCl effect, and Q designates a quadratic zinc effect. The superscript \* indicates a significant effect ( $P \leq 0.05$ ).