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The effects of feeding total mixed ration with high roughage content on growth performance, carcass characteristics, and meat quality of Hanwoo steers

Running title: High roughage fed Hanwoo steers' carcass characteristics and meat quality

Abstract

This study investigated the dietary effect of total mixed ration (TMR) based on high roughage content on the growth performance, carcass characteristics, and meat quality of Hanwoo steers. Twenty-four Hanwoo steers (average body weight, 195.3 ± 4.7 kg; age, 8.5 mon) were randomly allocated to three experimental groups according to forage and concentrate ratio (DM basis): 25:75 (control), 50:50 (T₅₀), and 70:30 (T₇₀). Productivity in the fattening period and final body weight were significantly higher in the control. Average daily gain and feed conversion ratio were the same among treatments. Serum parameters, cholesterol, blood urea nitrogen, and total protein were higher in the control. Carcass weight was comparable in the control and T₅₀ but feeding more roughage was significantly correlated with a higher intramuscular fat. Shear strength and drip loss were higher while n-6/n-3 was lower in T₇₀ compared to the other groups. However, meat color was not significantly different among treatments. In terms of free amino acid contents, glutamic acid and glycine were higher in the control than T₅₀ and T₇₀. Overall, feeding Hanwoo steers with high forage content TMR had the lowest n-6/n-3 ratio of fatty acid content but highest intramuscular fat, shear strength, and drip loss. High forage content TMR is the best feed for Hanwoo steers that gives more benefits

35 for human health and consumption but also provides the best meat grade and quality, which is
36 important in the beef market in Korea.

37

38 **Key words** Hanwoo steers, meat quality grade, roughage, total mixed ration

ACCEPTED

39 **Introduction**

40 Research on the productivity and utility of roughage in the beef cattle industry is
41 increasingly important. However, there is little domestic roughage production, and there have
42 been few studies on production, or on the productivity or product quality of cattle fed roughage.
43 Hence in South Korea, Hanwoo farms have recently expanded their roughage cultivation area
44 to self-produce roughage and reduce feed cost. In Jeollanam-do, the roughage cultivation area
45 increased from 32,000 ha in 2011 to 49,000 ha in 2019; 62% of the total production area was
46 for Italian rye grass (IRG), which increased from 13,757 ha in 2011 to 27,474 ha in 2019. In
47 this way, high-quality roughage is being produced based on domestic roughage demand,
48 reducing the need for imported feed ingredients.

49 In ruminants, roughage is essential for the healthy development of the rumen, and the
50 quality of roughage greatly affects productivity (Maeng et al., 1989). In South Korea, total
51 mixed ration (TMR) has been widely used for dairy cattle, and was introduced to Hanwoo in
52 the 2000s. Kim et al. (2003) and Li et al. (2003) investigated the effects of hay-based high-
53 fiber mixed feed on intraruminal fermentation properties, digestion coefficient, and late
54 fattening-stage growth of Hanwoo steers. Hanwoo cattle fed a mixture of rice straw and grass
55 exhibited an increased growth rate and feed intake than cattle fed only rice straw (Ahn et al.,
56 1984; Cho et al., 1997), whereas no major effect on carcass characteristics was observed (Cho
57 et al., 1997). Cho et al. (2008) investigated the effects of high-fiber mixed feed on weight gain,
58 carcass characteristics, and production cost of Hanwoo steer, and suggested that mixed feed
59 may be superior to conventional rice straw-based rearing methods. Moreover, studies on the
60 value-as-feed of soiling crops and silage in not only Hanwoo steer (Cho et al., 2000) but also
61 wool sheep (Lee et al., 2002) and black goats (Hwang et al., 2008), reported positive results in
62 terms of utility.

63 Whole-crop barley (WCB) and Italian ryegrass (IRG) are two of the of the most

64 preferred roughages for ruminants especially for beef producers. WCB silage has long been
65 proposed as a roughage ingredient to replace imported grains, and its use has resulted in
66 significantly increased daily weight gain and improved yield grade, carcass quality, backfat
67 fitness, and intramuscular fat compared with rice straw (Seo et al., 2010; Kook et al., 2011).
68 On the other hand, IRG has been also preferred forage crop due to high forage yield and good
69 nutritional quality. Kim et al. (2015) also reported that feeding IRG silage combined with
70 concentrate significantly increased crude fat and lightness (L*) of Hanwoo beef as well as
71 increase the rib-eye area, back fat thickness, and slaughter weight of the carcass trait.

72 In the growing and fattening period of beef steers, the forage level in diets influences
73 the meat production and meat quality (Sung et al., 2015). Angus steers fed with wet distillers
74 grain inclusion in high forage diet increases intramuscular fat content of the beef (Schoonmaker
75 et al., 2010). Also, high-roughage diets, especially those containing high-quality roughage,
76 produce meat with a deeper red color (Bidner et al., 1986). Among different types of roughage,
77 beef cattle fed grass silage exhibited a better meat color than those fed maize silage (O'Sullivan
78 et al., 2002). Thus, this present study examined the effects of TMR feeding using mostly high-
79 quality roughage on the growth performance, carcass characteristics, and meat quality of
80 Hanwoo steers.

81 **Materials and Methods**

82 **Experimental animals, treatments, feed and feeding management**

83
84 A 615-day rearing experiment was conducted in the testing cowsheds at the Livestock
85 Research Center of the Jeollanam-do Agricultural Research and Extension Services (Gangjin-
86 gun, Jeollanam-do, South Korea). We used 24 castrated Hanwoo steer calves (mean age, 8.5
87 mon; 195.3 kg \pm 4.7 kg), and prepared feed for four different growth stages (rearing, 8.5–12
88

89 mon; early fattening, 13–18 mon; mid-fattening, 19–24 mon; late fattening, 25–32 mon). There
90 were a total of three groups defined by the feeding plan at each growth stage: 1) a conventional
91 feeding group (control) were fed 25% roughage as specified by the National Institute of Animal
92 Science (2012), 2) a group fed 50% roughage on average throughout the whole rearing period
93 (T_{50}), and 3) a group fed 70% roughage on average (T_{70}). The animals were divided into three
94 pens with eight animals in each treatment group. In addition, when allocating the calves to the
95 pens, age and body weight were considered to reduce the differences in mean body weight
96 between the pens and minimize uneven feed intake due to dominance hierarchies.

97 A TMR feed was provided to each group that included IRG, maize silage, and barley
98 grain were produced by the Livestock Research Center. Other ingredients were purchased from
99 Samjeong Natural Co., Ltd. (TMR feed factory, Yeongam, Jeollanam-do, South Korea), and
100 prepared at the same factory in accordance with this study's feeding program (Hanwoo board,
101 2009; National Institute of Animal Science, 2012). The feed mixture ratios and chemical
102 composition for each group and growth stage are shown in Table 1. The levels of general
103 components in the feed were analyzed according to the AOAC (2000) method, and neutral
104 detergent fiber (NDF) and acid detergent fiber (ADF) were analyzed according to the method
105 of Van Soest et al. (1991). Feed was provided twice per day, at 8.30 AM and 4.30 PM, and the
106 animals were allowed to feed *ad libitum*. Water and mineral blocks (Super Licks, UK) were
107 available to all groups at all times.

108

109 **Body weight and blood composition**

110 Body weight was measured before the morning feeding at 2-mon intervals from
111 immediately before the start of the experiment to the end of the experiment. Daily weight gain
112 was calculated by dividing the difference between the previous weight measurement and the
113 current weight measurement by the number of days. Feed intake was calculated using a feed

114 intake recording system (Dawoon Electronics Co., Ltd, Incheon, South Korea), and the feed
115 conversion ratio was calculated by dividing the weight gain by the feed intake.

116 From the start to the end of the experiment, at the same time as weighing, 10 ml of
117 blood was collected from the jugular vein of each animal in a serum vacutainer (BD Vacutainer
118 Serum Tubes [REF 367820], Oakville, ON, USA), centrifuged for 15 min at 4°C and 2,000 ×
119 g (Hanil Science Co., Ltd, KRI supra 21 K, Daejeon, South Korea), and stored at -70°C in an
120 ultra-low temperature upright freezer (Thermo Scientific, Beverly, MA, USA) until analysis.
121 Serum glucose, cholesterol, creatinine, BUN, and total protein were measured using an
122 automated biochemistry analyzer (CIBA-EXPRESS PLUS, Ciba-Corning Diagnostics, MA,
123 USA).

124

125 **Carcass analysis**

126 For carcass analysis, after the feeding experiment ended, the experimental animals were
127 slaughtered by Seorim Global Co., Ltd. (Damyang, Jeollanam-do, South Korea). The carcasses
128 were hung for 18–24 h at 0°C, and then the meat yield (carcass weight, backfat thickness, and
129 cross-sectional area of longissimus dorsi) and meat quality (intramuscular fat [marbling], meat
130 color, fat color, texture, and maturity) factors were rated by a livestock grader in accordance
131 with the Carcass Grading System for Cattle.

132

133 **Meat quality characteristics**

134 The general composition of Hanwoo meat samples (sirloin) was analyzed in accordance
135 with AOAC (2000) methods. First, 3 g of pulverized sample was dried for 24 h in a 104°C dry
136 oven, and the water content was determined by comparing the weight before and after drying.
137 Protein was measured using a Kjeltex System (Kjeltex Auto 2400/2460, Foss Tecator AB,
138 Höganäs, Sweden), fat content was measured using the Soxhlet method, and ash content was

139 measured using an ash analyzer (MAS 70-00, CEM Corp., Matthews, NC, USA).

140 In the analysis of physicochemical characteristics, pH was measured by homogenizing
141 2 g of sample in 18 mL of distilled water with a homogenizer (Polytron PT 10-35 GT,
142 Kinematica, Lucerne, Switzerland) at $15,800 \times g$ for 30 s, passing the solution through a
143 Whatman No. 4 filter paper, and measuring the filtrate with a pH meter (Orion 2-Star, Thermo
144 Scientific). For water holding capacity, 5 g of ground sample was collected, placed in a 50-mL
145 tube with cotton wool, and centrifuged for 10 min at $112 \times g$ before weighing. To ascertain loss
146 on heating, a 2-cm-long sample was cut and weighed, before placing in a vacuum pack and
147 heating at a constant temperature of 80°C . When the core temperature of the sample reached
148 70°C (~8 min), the sample was cooled with cold water and then weighed. To measure shear
149 strength, holes were made using a 1-cm-diameter cork in the samples from the loss-on-heating
150 test. After fitting a Warner-Bratzler blade to a texture analyzer (TA-XT2, Stable Micro
151 Systems, Surrey, UK), the blade was positioned perpendicular to the direction of the muscle
152 fibers, and the shear strength (kg) was measured. The device conditions were set to pre-test
153 speed 2.0 mm/s, test speed 2.0 mm/s, and post-test speed 5.0 mm/s. To measure drip loss, a 1-
154 cm piece of sample was cut and weighed, placed in oxygen-permeable paper, stored for 48 h
155 in a 4°C refrigerator, and then weighed. To measure surface meat color, three locations on the
156 sample surface were arbitrarily selected, and a color meter (Model CR-410, Minota Co. Ltd.,
157 Osaka, Japan) was used to measure the CIE L^* (lightness), CIE a^* (redness), and CIE b^*
158 (yellowness). As a reference color, a white plate was used with L^* 89.2, a^* 0.921, and b^* 0.783.

159 To analyze the fatty acid composition, 1 g of sample was mixed with 0.7 mL of 10 N
160 KOH in water, and placed in a thermostatic water bath preheated to 55°C . While heating for 1
161 h 30 min, the mixture was vigorously stirred once every 30 min. Next, the mixture was cooled
162 for 1–2 min and then heated for another 1 h 30 min in the 55°C water bath while being
163 vigorously stirred once every 30 min. After heating, the mixture was cooled in cold water,

164 before adding 3 mL of hexane and centrifuging for 5 min at $1,008 \times g$. After using a Pasteur
165 pipette to transfer the mixture to a vial, a gas chromatograph-flame ionization detector (Agilent
166 Technologies, 7889 Series, Santa Clara, CA, USA) was used to analyze the fatty acid
167 composition. To measure the free amino acid content, the method of Hughes et al. (2002) was
168 used to extract free amino acids. After adding 10 mL of 2% TCA solution to 2.5 g of finely
169 pulverized sample, homogenization was performed for 1 min at $20\,412 \times g$ min. The
170 homogenate was centrifuged for 15 min at $17,000 \times g$, filtered using a 0.45- μ m membrane
171 filter, and derivatized by the Waters AccQ-Tag method (1993, Waters Corporation, Milford,
172 MA, USA) to make the free amino acid sample, which was measured using RP-HPLC. The
173 column used here was an AccQ-Tag column (3.9×150 mm, Waters), the injected volume was
174 5 μ L, column temperature was 37°C, detector was a fluorescent detector (Waters 2475,
175 Millipore Co-operative, Milford, MA, USA), excitation wavelength was 250 nm, and emission
176 wavelength was 395 nm. A gradient method was used for analysis with mobile phase solvents
177 of Waters AccQ-Tag eluent A (solvent A) and 60% acetonitrile (solvent B).

178

179 **Statistical analysis**

180 All the results obtained in the present study were analyzed using the SAS package
181 (Statistical Analysis System, 2003). Duncan's multiple range test was used to compare means
182 between the groups at a significance level of 5%.

183

184

185 **Results and Discussion**

186

187 **Growth characteristics**

188 The effects of TMR feeding using high-quality roughage (IRG, maize silage, and barley
189 grain) on weight gain characteristics of Hanwoo steers were investigated and compared with

190 those for conventional feeding. The total weight gain, daily weight gain, and feed conversion
191 ratio of the Hanwoo steer at each growth phase are shown in Table 2. For Hanwoo steer calves
192 with a mean weight of 195.3 kg, during the 105-day rearing stage, the total weight gain was
193 79.0 kg in the control group, 76.4 kg in T₅₀, and 80.2 kg in T₇₀; the total weight gain was
194 significantly higher in T₇₀ than in the control group ($p<0.05$), and there were no significant
195 differences between the groups in terms of end weight, daily weight gain, or feed conversion
196 ratio. During the 150-day early fattening stage, the total weight gain was 135.0 kg in the control
197 group, 123.8 kg in T₅₀, and 112.1 kg in T₇₀, which was significantly higher in the control group
198 ($p<0.05$), but there were no differences between the groups in terms of feed conversion ratio.
199 During the 150-day mid-fattening stage, total weight gain was higher in the control group
200 (conventional feeding; $p<0.05$), but there were no differences in daily weight gain or feed
201 conversion ratio. During the 210-day late fattening stage, total weight gain was 176.0 kg in the
202 control group, 172.3 kg in T₅₀, and 185.9 kg in T₇₀, meaning that the highest total weight gain
203 was in T₇₀ ($p<0.05$). This result was considered to be the result of compensatory growth, since
204 the ratio of concentrates in the early and mid-fattening stages (25–40%) was low in T₇₀, but the
205 ratio of concentrates increased in the late fattening stage (60%). Overall, a high proportion of
206 forage in TMR resulted in lower rate of growth across the whole experimental period. This
207 result is in agreement with Thomas et al. (1988) and Baker et al. (1992) who reported that cattle
208 fed diets containing a higher proportion of grass silage sustained a lower growth rate than cattle
209 given high-concentrate diets.

210

211 **Carcass analysis**

212 Table 2 shows the live weight, carcass weight, backfat characteristics, meat yield, and
213 meat quality characteristics measured after the feeding experiment was completed. In terms of
214 live weight, the groups fed a high-quality roughage-based diet had lower mean body weight

215 than the control group (conventional diet) ($p < 0.05$), and carcass weight exhibited a similar
216 pattern with lower weights observed in the treatment groups than the control group ($p < 0.05$).
217 These results can be considered to reflect the weight gain during the feeding experiment. On
218 the other hand, the sirloin cross-sectional area and intramuscular fat (Table 2) had comparable
219 results in T₅₀ and T₇₀ and both significantly higher in intramuscular fat than the control group
220 ($p < 0.05$). However, T₅₀ was also significantly higher in the sirloin cross-sectional area than the
221 control group ($p < 0.05$), whereas there no significant change in backfat thickness or meat yield
222 index was found. This study corroborates with Sung et al. (2015) study on Hanwoo steers fed
223 with high forage diet and high forage diet with chromium methionine that tended to show and
224 significantly higher intramuscular fat than low forage diet and low forage diet with chromium
225 methionine, respectively. They also explained that the precursor of intramuscular fat is the
226 glucose that usually from the high ruminal starch digestion, which increases the organic acids
227 that are later converted to glucose. This indicates that high roughage diet in this study contains
228 high quality roughages that was converted to glucose that produced significantly high
229 intramuscular fat. Kim et al. (2015) reported that the carcasses of Hanwoo steers fed with an
230 IRGS/concentrate diet resulted in an increase in backfat thickness and rib-eye area compared
231 with those fed with rice straw/concentrate diet. Our study is in agreement with their result for
232 cross-sectional area of sirloin; however, no significant change in the backfat thickness was
233 observed in our study. However, live weight and carcass weight were reduced in steers fed with
234 high forage content (T₇₀). Our result is in agreement with the findings of Steen (1995), who
235 found that the carcass weight of steers reduced when concentrates intake was reduced.

236

237 **Blood characteristics**

238 Table 3 shows the changes in serum concentration of metabolites in Hanwoo steer by
239 growth stage depending on the ratio of roughage in their diet. Serum cholesterol was higher in

240 the control group in the mid- and late fattening stages ($p < 0.05$), and BUN and total protein
241 were higher in the early and late fattening stages ($p < 0.05$). Metabolites in the blood are
242 indicators that enable measurement of the use and metabolism of nutrients (Vernon, 1992; Choi
243 et al., 2009). Cholesterol concentration is positively correlated with energy intake (Arave et al.,
244 1975), and the higher cholesterol levels in the control group in the fattening stages was thought
245 to be due to the higher intake of concentrates relative to the treatment groups. Enright et al.
246 (1990) and Choi et al. (2009) reported that decreased serum BUN levels indicate the
247 accumulation of nitrogen in tissues engaged in protein synthesis. The BUN levels from the
248 present study were in the normal range of 10–20 mg/dl (Kwon et al., 2005; Choi et al., 2006).
249 Therefore, the differences between the groups are thought to be due to the type of feed and
250 differences between individual animals (Choi et al., 2006, 2009).

251

252 **Meat quality characteristics**

253 When six animals were analyzed in each group, water content, crude protein, crude fat,
254 and crude ash content in Hanwoo sirloin were not affected by the amount of roughage in the
255 feed. These results are shown in Table 4 alongside the pH, water holding capacity, loss on
256 heating, shear stress, and drip loss. The shear strength value and drip loss of Hanwoo steers fed
257 TMR with high forage content (T₇₀) were significantly higher than that of other cattle ($p < 0.05$).
258 However, previous studies reported no significance in pH, WHC, cooking loss, shear strength,
259 or drip loss with changes in forage content. Furthermore, our results are in disagreement with
260 the study of Frank et al. (2016), who reported that meat with high IMF has lower drip loss and
261 water loss. The muscular condition of experimental animal rigor mortis affected the shear
262 strength and drip loss when fed with TMR with high forage content. pH is closely related to
263 changes in quality, such as water holding capacity and tenderness, and so is fundamental to
264 assessing the quality of meat (Weatherly et al., 1998). Water holding capacity refers to the

265 ability of meat to retain initial or added water content when subjected to physical forces such
266 as shear stress, pulverization, pressure, or heat. Water holding capacity affects meat color,
267 texture, hardness of fresh meat, tenderness of cooked meat, and juiciness, and water holding
268 capacity has been reported to increase with changes in the protein structure and ionic strength
269 of meat (Wu and Smith, 1987). According to Laster et al. (2008) and Obuz et al. (2004), shear
270 strength, which reflects tenderness, decreases with increasing meat quality grade, which is
271 contrary to the results of the present study. Drip loss is caused by changes in the microscopic
272 structure of muscle due to sarcomere contraction; as muscle contracts, the space inside the
273 muscle decreases, and water leaks out of the muscle, resulting in weight loss (Kim et al., 1994).

274 Meat color is determined by several factors, but is most strongly influenced by the
275 concentration and chemical state of myoglobin inside the muscle. Meat color darkens with
276 increased age, and also differs depending on feed and species. The lightness, redness, and
277 yellowness of sirloin were measured, and none showed any differences between the groups
278 (Table 4). Therefore, increasing the ratio of roughage in feed had no major effect on meat color.
279 The fatty acid composition showed no significant differences between groups, and so was
280 believed to be unaffected by the ratio of roughage (Table 5). As anticipated, the ratio of oleic
281 acid (46.94–47.42%) was the highest, followed by palmitic acid (24.43–25.34%) and stearic
282 acid (11.31–11.59%). The ratio of saturated fatty acid (SFA) among all fatty acids was 39.80–
283 39.03% in all groups, and the ratio of unsaturated fatty acid (UFA) was 55.67–56.41%.
284 Mahecha et al. (2009) reported that the ratio of SFA:UFA in beef was 47.53:52.47, representing
285 a higher ratio of SFA and lower ratio of UFA compared with the results of the present study.
286 In addition, the ratio of monounsaturated fatty acid among UFA was 52.68–53.05%, and the
287 ratio of polyunsaturated fatty acid was 2.91–3.04%. The UFA/SFA ratio in Hanwoo steer has
288 been found to be in the range 1.41–1.45. The contents of linoleic acid (C18:2) and linolenic
289 acid (C18:3) of steers fed with a high forage content increased and decreased, respectively. Our

290 results show that steers fed with a high forage content TMR had a lower n-6/n-3 value, which
291 is in agreement with the results of Wood et al. (1999). Wood et al. (1999) also stated that
292 feeding grass or grain diets is important in changing the n-6/n-3 value; therefore, feeding high
293 roughage diet TMR in Hanwoo steers favors low n-6/n-3 value. The high n-6/n-3 value
294 promotes the heart disease and cancer (Kang, 2004); hence, low n-6/n-3 value in Hanwoo steers
295 fed with a high forage content TMR gives more benefits for human health and consumption by
296 decreasing the risk of heart disease and cancer.

297 The free amino acid concentration, which affects the savory and sweet tastes of beef,
298 showed lower glutamic acid and glycine ($p < 0.05$) in steers fed with a high forage content diet.
299 Other amino acids were observed at higher levels in the control group, but there were no
300 significant differences according to the ratio of roughage in the diet (Table 6). The free amino
301 acid content in the carcasses of feeder cattle has been reported to be affected by the amino acid
302 content of the feed ingredients (Kim et al., 2011). The free amino acids related to beef taste are
303 caused by the hydrolysis of cooked free amino acids and reaction with lipid oxidation products,
304 and is reported to be affected by the initial state of the meat (Shibamoto, 1980). However, the
305 free amino acid concentration in beef is not the only factor affecting its taste, and taste is known
306 to be produced by interactions between amino acids, sugars, and fatty acids during heating
307 (Macleod, 1994). Sirloin from Hanwoo steer fed a high-roughage diet (T₇₀) exhibited slightly
308 higher scores for meat color and tenderness, but the other variables showed no significant
309 differences between the treatment groups and the control group.

310

311 **Conclusion**

312 Feeding of TMR with high forage content increased the shear strength and drip loss of
313 the carcasses. Moreover, the intramuscular fat or the intramuscular fat was higher in TMR with
314 50:50 and 70:30 forage and concentrate ratio than control. Also, steers fed TMR with high

315 forage content the lowest n-6/n-3 ratio. Lowest n-6/n-3 ratio of fatty acid content but highest
316 intramuscular fat in steers fed with high forage content TMR is more beneficial for human
317 health and consumption as well as for the Hanwoo producers.

318

319 **Conflicts of Interest**

320 The authors declare no potential conflicts of interest.

321

322 **Author contributions**

323 Conceptualization: Ku MJ; Data curation: Ku MJ, Choi YS; Formal analysis: Mamuad LL, Lee
324 SS; Methodology: Ku MJ, Choi YS, Nam KC, Cho YI; Software: Ku MJ, Choi YS, Mamuad
325 LL, Nam KC; Validation: Ku MJ, Choi YS, Kim SS, Mamuad LL, Lee SS; Investigation: Ku
326 MJ, Mamuad LL, Kim SS, Nam KC, Cho YI, Lee SS; Writing - original draft: Ku MJ, Mamuad
327 LL, Lee SS; Writing - review & editing: Ku MJ, Mamuad LL, Nam KC, Cho YI, Lee SS.

328

329 **Ethics Approval**

330 The experimental protocol was approved by the Institutional Animal Care and Use
331 Committee at the Livestock Research Institute, Jeollanamdo Agricultural Research & Extension
332 Service (JARES) (JLRI-IACUC 2015-02).

333

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338

339

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465 Table 1. Feed mixture ratios and chemical composition (%) of Hanwoo steers fed with conventional (control; 25% roughage), 50% roughage (T₅₀),
 466 and 70% roughage (T₇₀) diets by growth stage (dry weight, %)

Composition	Growing (8.5–12 mon)			Early fattening (13–18 mon)			Mid-fattening (19–24 mon)			Late fattening (25–32 mon)		
	Control	T ₅₀	T ₇₀	Control	T ₅₀	T ₇₀	Control	T ₅₀	T ₇₀	Control	T ₅₀	T ₇₀
Feed mixture ratios												
Alfalfa	5.18	3.27	-	-	-	-	-	-	-	-	-	-
Rice straw	-	-	-	12.8	17	-	8.4	14	-	6	7.8	6.8
Corn silage	12.96	21.8	32.3	14	22	24	10.4	10	22	-	-	-
Italian rye grass (dry)	32.98	36.6	48	18.3	25	56	16.7	16	34	4.1	20	34
Barley	8.25	5.25	3.92	5.5	6.1	4	6.2	6	6	8.6	5.8	3.4
Corn gluten feed	4.48	4.16	1.96	6	3.3	1.2	7.7	6.5	3.8	12	11	10
Molasses	5.54	5.25	2.94	2.6	2.4	2.4	2.8	3.3	2.2	4.4	3.4	3
Wheat bran	4.48	4.16	1.96	6	3.3	1.2	5.6	4.4	1.8	4.2	4.8	3.2
Coconut meal	2.24	1.58	0.98	3	1.7	0.6	2.8	2.2	0.9	1.5	1.5	1
Corn flakes	3.06	3.07	-	18.3	11	9.2	18.2	22	16	29.9	27	25
Cottonseed	-	-	-	-	-	-	4.2	3.3	4	4	3.9	2.7
Apple pomace	1.06	0.99	0.69	1.5	0.8	0.3	1.3	1	0.4	0.7	0.7	0.5
Rice bran	8.25	4.95	2.94	5.5	5	8	6.3	5	5	8.2	5.8	2.7
Palm kernel meal	1.18	1.39	0.49	2	1	0.4	1.9	1.5	0.6	1	1	0.7
Water	9.42	6.44	2.94	3	2.2	2	5.2	3.3	2	13.1	5.8	6
Vitamin and mineral supplement	0.24	0.3	0.2	0.3	0.3	0.3	0.3	0.3	0.3	0.2	0.2	0.2
Limestone	0.59	0.59	0.59	1.1	0.7	0.6	0.6	0.7	0.6	0.9	0.8	0.6
Salt	0.12	0.2	0.1	0.2	0.1	0	0.2	0.1	0.1	0.2	0.1	0.1
Chemical composition (%)												
Water	36.7	44.8	41.3	24.1	35	39.5	34.8	33.1	36.9	24.1	23.5	22.6
Crude protein	11.8	10.4	10.1	9.9	7.7	6.5	8.6	8.1	7.5	9.6	8.7	7.5
Crude fat	2.9	2.5	3.7	4.2	3.4	3	3.5	3.4	3.1	3.8	3.3	3
Crude fiber	15.7	13.7	15.8	22.1	16	15.2	13.4	14.7	15.3	17.3	16.9	15.1
Crude ash	7	7.4	7.3	10	10.3	8.9	5.7	5.8	6.3	6.1	6.5	6.2
Calcium	0.8	0.7	0.8	1.3	0.6	0.5	0.4	0.5	0.5	0.6	0.6	0.5
Phosphorus	0.3	0.3	0.4	0.4	0.3	0.3	0.3	0.4	0.3	0.3	0.4	0.3
Neutral detergent fiber	31.3	33.2	37.2	42.4	38.6	33	33.8	34.2	33.2	36.4	36.6	40.9
Acid detergent fiber	16.1	15.6	17	24.5	24.7	20.2	20.9	19.7	20.6	23.9	19.8	22.9
Total digestible nutrient	59.9	54.9	56.1	61.2	59.1	59.1	63.8	65.3	60.7	71.5	70.1	69.8

468 Table 2. Steer productivity characteristics and carcass outcomes of Hanwoo steers fed with
 469 conventional (control; 25% roughage), 50% roughage (T₅₀), and 70% roughage (T₇₀) diets

Item	Control	T ₅₀	T ₇₀	SEM ¹⁾
Steer productivity characteristics				
Start weight (kg)	536.9 ^a	521.5 ^{ab}	505.1 ^b	12.532
End weight (kg)	712.9 ^a	693.8 ^b	691.0 ^b	13.654
Total weight gain (kg)	176.0 ^b	172.3 ^b	185.9 ^a	4.347
Daily weight gain (kg)	0.84	0.82	0.89	0.039
Feed conversion ratio	17.90	16.45	15.25	0.762
Meat yield characteristics				
Live weight (kg)	712.9 ^a	693.8 ^b	691.0 ^b	13.654
Carcass weight (kg)	409.6 ^a	400.9 ^a	392.9 ^b	15.401
Backfat thickness (mm)	10.7	11.3	14.0	3.982
Cross-sectional area of sirloin (cm ²)	78.1 ^b	84.4 ^a	79.3 ^{ab}	8.345
Meat yield index	65.0	65.7	63.5	2.701
Meat yield grade (A:B:C, %)	0:100:0	12:76:12	25:50:25	-
Carcass grading characteristics				
Intramuscular fat	5.7 ^b	6.4 ^a	6.0 ^a	1.996
Meat quality grade (1 ⁺⁺ :1 ⁺ :1:2,%)	28:29:14:29	13:62:25:0	38:25:12:25	-

470 ¹⁾Standard error of the means (n = 6)

471 ^{a,b}Values with different letters differ significantly (p<0.05).

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477 Table 3. Blood characteristics of Hanwoo steers fed with conventional (control; 25% roughage), 50%
 478 roughage (T₅₀), and 70% roughage (T₇₀) diets by growth stage

Item	Period	Control	T ₅₀	T ₇₀	SEM ¹⁾
Glucose (mg/dL)	Growing	63.27	62.50	63.98	3.056
	Early fattening	60.27	61.83	64.56	2.899
	Mid-fattening	72.35	73.15	75.17	2.571
	Late fattening	60.17	66.04	64.50	3.254
Cholesterol (mg/dL)	Growing	156.6	148.0	146.2	6.781
	Early fattening	189.3	149.1	136.0	6.248
	Mid-fattening	129.3 ^a	104.8 ^{ab}	98.5 ^b	5.893
	Late fattening	160.4 ^a	104.3 ^{ab}	92.2 ^c	6.735
Creatine (mg/dL)	Growing	1.00	0.86	0.88	0.054
	Early fattening	1.32	1.25	1.12	0.076
	Mid-fattening	0.98	0.92	0.87	0.069
	Late fattening	1.27	1.23	1.16	0.078
BUN ² (mg/dL)	Growing	11.34	11.98	11.76	0.765
	Early fattening	13.53 ^a	9.55 ^{ab}	8.62 ^b	0.585
	Mid-fattening	8.62	5.89	7.21	0.693
	Late fattening	14.56 ^a	8.63 ^{ab}	7.27 ^b	0.791
Total protein (g/dL)	Growing	11.21	11.76	11.98	0.107
	Early fattening	13.51 ^a	9.58 ^{ab}	8.69 ^b	0.121
	Mid-fattening	8.43	7.22	6.61	0.092
	Late fattening	11.20 ^a	8.97 ^{ab}	7.56 ^b	0.106

479 ¹⁾Standard error of the means (n = 8)

480 ^{a,b}Values with different letters differ significantly (p<0.05)

481 ²Blood urea nitrogen

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483 Table 4. General components, meat quality properties, and meat color of loins from Hanwoo steers
 484 fed with conventional (control; 25% roughage), 50% roughage (T₅₀), and 70% roughage (T₇₀) diets

Item	Control	T ₅₀	T ₇₀	SEM ¹⁾
Water content (%)	60.06	58.11	58.80	3.723
Crude protein (%)	21.03	20.24	21.44	2.128
Crude fat (%)	18.53	22.13	20.80	1.357
Crude ash (%)	0.93	0.94	0.85	0.052
pH	5.61	5.66	5.59	0.035
Water holding capacity (%)	76.69	75.26	74.29	5.189
Loss on heating (%)	13.30	13.38	13.85	2.592
Shear strength (kg.f)	1.77 ^b	1.91 ^a	1.93 ^a	0.064
Drip loss (%)	26.37 ^{ab}	24.78 ^b	28.46 ^a	2.018
L value	42.50	42.70	42.95	2.295
a value	27.28	27.38	27.00	1.593
b value	13.13	13.34	13.11	1.231

485 Standard error of the means (n = 6)

486 L : lightness, a: redness, b : yellowness

487 ^{a,b}Values with different letters differ significantly (p<0.05).

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490 Table 5. Fatty acid composition (%) of Hanwoo loins from steers fed with conventional (control; 25%
 491 roughage), 50% roughage (T₅₀), and 70% roughage (T₇₀) diets

Item	Control	T ₅₀	T ₇₀	SEM ¹⁾
Capric acid (C10:0)	0.03	0.03	0.03	0.000
Lauric acid (C12:0)	0.07	0.07	0.06	0.002
Myristic acid (C14:0)	2.76	2.85	2.83	0.081
Myristoleic acid (C14:1)	0.78	0.92	0.87	0.074
Palmitic acid (C16:0)	25.28	24.43	25.34	0.258
Palmitoleic acid (C16:1)	3.77	3.86	3.91	0.101
Heptadecenoic acid (C17:1)	0.63	0.65	0.62	0.088
Stearic acid (C18:0)	11.59	11.56	11.31	0.150
Oleic acid (C18:1)	46.98	46.94	47.42	0.349
Linoleic acid (C18:2)	1.91	1.86	1.67	0.917
Linolenic acid (C18:3)	0.73	0.82	0.86	0.052
Arachidic acid (C20:0)	0.07	0.07	0.07	0.000
Arachidonic acid (C20:4)	0.33	0.32	0.25	0.022
SFA ²⁾	39.80	39.00	39.03	0.338
UFA ³⁾	55.67	56.08	56.41	0.369
MUFA ⁴⁾	52.68	53.05	52.88	0.322
PUFA ⁵⁾	2.99	3.04	2.91	0.024
UFA/SFA	1.41	1.45	1.41	0.019
n-6/n-3	2.63 ^a	2.29 ^a	1.98 ^b	0.166

492 ¹⁾Standard error of the means (n = 6)

493 ²⁾Saturated fatty acid

494 ³⁾Unsaturated fatty acid

495 ⁴⁾Monounsaturated fatty acid

496 ⁵⁾Polyunsaturated fatty acid

497 Table 6. Free amino acid composition (mg/100 g) of Hanwoo loins from steers fed with conventional
 498 (control; 25% roughage), 50% roughage (T₅₀), and 70% roughage (T₇₀) diets

Item	Control	T ₅₀	T ₇₀	SEM ¹⁾
Alanine	38.94	34.56	26.20	2.912
Arginine	5.84	5.34	5.29	0.081
Asparagine	13.28	12.65	10.59	1.168
Aspartic acid	3.42	2.86	2.44	0.766
Cystine	9.77	8.92	6.80	0.654
Glutamic acid	30.62 ^a	19.23 ^{ab}	6.79 ^b	2.789
Glycine	21.36 ^a	18.92 ^a	13.14 ^b	2.125
Histidine	6.44	6.22	4.22	0.823
Isoleucine	7.61	6.83	5.87	0.890
Leucine	20.90	19.16	16.06	3.112
Lysine	8.36	7.88	6.86	1.598
Methionine	0.34	0.25	0.33	0.002
Phenylalanine	11.45	10.12	7.94	1.669
Proline	2.16	2.03	1.58	1.011
Serine	13.10	12.44	10.00	1.768
Taurine	15.39	14.35	14.09	1.562
Threonine	8.04	7.76	6.27	0.993
Tryptophan	10.68	9.14	9.74	1.540
Tyrosine	10.80	10.05	8.29	1.898
Valine	9.83	9.15	7.86	1.161

499 ¹⁾Standard error of the means (n = 6)

500 ^{a,b}Values with different letters differ significantly (p<0.05).

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