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# The optimum grazing time based on milk fatty acids and yield for Holstein lactating cows through eco-pastoral system in alpine grassland

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**Key words:** Holstein cow; Milk fatty acids; Milk yield; Optimum grazing time; Eco-pastoral system

## Abstract

This study aimed to detect the optimum grazing time based on milk fatty acids and yield for the Holstein cow (*Bos taurus*) through eco-pastoral system in Korean alpine grassland. The treatment of grazing time was 3 levels: Non-grazing, Half-day grazing, Full-day grazing. First, the difference of milk fatty acids (saturated fatty acid, unsaturated fatty acid, Total  $\omega$ -3, etc.) and milk yield was checked by grazing time via ANOVA. Second, various measurements of milk fatty acids were reduced to principal components for effective and obvious detecting the variation as a grazing time. Final, the variations of milk fatty acid and yield were estimated to calculate the optimum grazing time by regression modeling based on multinomial function. For detecting the optimum grazing time, checking the crossing point between increases of milk fatty acid and decreases of milk yield, and response surface methodology were carried out. As a result, the optimum grazing time was 11.05 h by response surface methodology, respectively. For the optimum grazing time, unsaturated fatty acid, mono-unsaturated fatty acid, eicosatrienoic acid, total  $\omega$ -3 and milk yield were 17.82 g/100g, 9.14 g/100g, 0.61 g/100g, 2.06 g/100g and 27.16 kg/d, respectively. The 11-hour grazing time was statistically optimal considering milk fatty acid and yield, but it would be minimal level if the loss due to milk yield reduction was covered by a premium grade and potential and long-term benefits were taken into account.

## Introduction

In the Republic of Korea, 64 % of the country is made up of mountainous area, and the area available for development as grassland is estimated at 1,711,000 ha, which is 26.6 % (MAFRA, 2016); however, the actual managed grassland are 34,000 ha. Nowadays the global price of feedstuffs is rising, making Korean farmers burdensome. Because the feedstuffs to import occupies more than 60 % of the domestic total production cost of animal husbandry. Sung et al. (2017) reported that the Korean standard model for utilization of pasture in alpine grassland that could overcome the drawbacks and promote eco-friendly grazing system. Furthermore, Lee et al. (2018) reported the difference in feed intake, milk yield (MY), quality, and fatty acid profile between mountainous grazing and feedlot systems for Holstein lactating cows.

Eco-pastoral system in alpine grassland is the method which utilizes the pasture in the mountainous areas through integrating livestock grazing system (Bueno et al., 2011). Grazing by the eco-pastoral system widely contributes to not only animal welfare, but also nature and human wellbeing. Furthermore, environmental change to livestock activities by grazing contributed to develop a natural system with interaction between grassland and herbivore in dynamic trade-off reflecting the eco-pastoral system (Austrheim and Eriksson, 2001). In particular, the eco-pastoral system could be called as ecosystem services, includes both ecological process and the benefits to human that can be derived from them (Balmford et al., 2008). Furthermore, the ecosystem services provided by mountainous area have contributed to provision of high quality water and foods, preservation against abnormal weather phenomena and biodiversity (World Bank, 2008). However, there is somewhat concern about a decline in MY in terms of economic feasibility. Sometimes the concern could be easily quantified relative to the merits, which might make the performance itself impossible at the planning step. In the Republic of Korea, the productivity by grazing considered to be low due to the lack of proper grazing practices and small size of alpine grassland for grazing (Sung et al., 2017). Furthermore, they pointed out the lack of knowledge of milk quality and milk fatty acid (MFA) profile of the Korean alpine grazing. Therefore, MFAs and MY were considered as a representative feature to find the optimum grazing time through eco-pastoral system in this study.

Therefore, this study aimed to detect the optimum grazing time considering MFAs and MY for Holstein cows through eco-pastoral system in alpine grassland.

## Methods and Study Site

The study was conducted at the Sky Ranch near Pyeongchang (latitude: 128°35', longitude: 37°33') of the Republic of Korea, on a mixed pasture (2.19 ha) with rolling topography and elevations (890-930 m) from May 21 to June 25 (36 days) in 2018. Vegetation is predominantly grasses (90 % timothy (*Phleum pratense* L.), 5 % orchardgrass (*Dactylis glomerata* L.) and 5 % white clover (*Trifolium repens* L.)). Holstein cows (n = 21) were selected based on similar characteristics contains weight, calving number, daily MY and days in milk before grazing. Three treatments were: (1) Non-grazing (NG, 0 h), (2) Half-day grazing (HG, 12 h), and (3) Full-day grazing (FG, 24 h). Where, cows of NG were fed the self-compounding total mixed ration (TMR) and concentrate.

The solutions of reagents consisted of internal standard 2 mL (Glyceryl triundecanoate 5 mg and Chloroform 10 mL) and 7 % Boron trifluoride-methanol solution (14% Boron trifluoride-methanol solution 10 mL and Methanol 20 mL). MFAs were saturated fatty acid (SFA, g/100g), unsaturated fatty acid (UFA, g/100g), mono-unsaturated fatty acid (MUFA, g/100g), poly-unsaturated fatty acid (PUFA, g/100g), linoleic acid (LA, g/100g),  $\gamma$ -linolenic acid ( $\gamma$ -LA, g/100g), homo  $\gamma$ -linolenic acid (HLA, g/100g), archidonic acid (AA, g/100g), total  $\omega$ -6 (TN6, g/100g),  $\alpha$ -linolenic acid ( $\alpha$ -LA, g/100g), eicosatrienoic acid (EA, g/100g), eicosa pentaenoic acid (EPA, g/100g), docosa hexaenoic acid (DHA, g/100g), total  $\omega$ -3 (TN3, g/100g), ratio  $\omega$ -6/ $\omega$ -3 (RN) and al index (AI, g/100g).

## Results

### *Differences in milk fatty acids and yield as a grazing time*

As a result of checking the difference of characteristics via one-way ANOVA (Table 1), the weight, MY and days in milk were not different before grazing ( $p > 0.05$ ). The difference in SFA, UFA, MUFA, EA, TN3, RN and AI between groups were significant in MFAs ( $p < 0.05$ ), and MY by grazing time was different ( $p < 0.05$ ). Therefore, there were not significant differences in MFA and MY between treatments before grazing.

Table 1. Difference of characteristics of Holstein cows before grazing, milk fatty acids and yield as a grazing time by analysis of variance (ANOVA) and Turkey post-hoc.

Variables	Treatments			F <sup>a)</sup> (p-Value)
	NG (n = 7)	HG (n = 7)	FG (n = 7)	
Weight (Kg)	616.09±17.98 <sup>a</sup>	612.65±19.68 <sup>a</sup>	658.14±19.28 <sup>a</sup>	0.90 (p = 0.42)
Milk yield before grazing (Kg/d)	36.87±3.08 <sup>a</sup>	31.04±3.19 <sup>a</sup>	29.69±1.35 <sup>a</sup>	2.03 (p = 0.16)
Days in milk (d)	166.43±8.52 <sup>a</sup>	183.14±11.69 <sup>a</sup>	187.57±14.33 <sup>a</sup>	1.78 (p = 0.20)
Saturated fatty acid (g/100g)	85.69±1.02 <sup>a</sup>	81.50±0.51 <sup>b</sup>	78.55±0.65 <sup>b</sup>	22.38*
Unsaturated fatty acid (g/100g)	14.31±1.02 <sup>a</sup>	18.50±0.51 <sup>b</sup>	21.45±0.65 <sup>b</sup>	22.38*
Mono-unsaturated fatty acid (g/100g)	5.90±1.14 <sup>a</sup>	10.42±0.35 <sup>b</sup>	11.86±0.74 <sup>b</sup>	14.60*
Poly unsaturated fatty acid (g/100g)	8.40±0.36 <sup>a</sup>	8.08±0.64 <sup>a</sup>	9.60±0.21 <sup>a</sup>	3.24 (p = 0.11)
Linoleic acid (g/100g)	3.50±0.36 <sup>a</sup>	2.32±0.20 <sup>a</sup>	3.73±0.53 <sup>a</sup>	3.81 (p = 0.09)
$\gamma$ -Linolenic acid (g/100g)	2.05±0.11 <sup>a</sup>	1.92±0.14 <sup>a</sup>	1.51±0.13 <sup>a</sup>	4.70 (p = 0.06)
Homo Linolenic acid (g/100g)	0.46±0.07 <sup>a</sup>	0.61±0.21 <sup>a</sup>	0.35±0.08 <sup>a</sup>	0.90 (P = 0.46)
Arachidon acid (g/100g)	0.76±0.15 <sup>a</sup>	1.08±0.12 <sup>a</sup>	1.02±0.11 <sup>a</sup>	1.77 (p = 0.25)
Total $\omega$ -6 (g/100g)	6.77±0.36 <sup>a</sup>	5.93±0.62 <sup>a</sup>	6.61±0.31 <sup>a</sup>	1.00 (p = 0.43)
$\alpha$ -Linolenic acid (g/100g)	0.17±0.08 <sup>a</sup>	0.17±0.05 <sup>a</sup>	0.42±0.10 <sup>a</sup>	3.36 (p = 0.11)
Eicosatrienoic acid (g/100g)	0.32±0.13 <sup>a</sup>	0.57±0.14 <sup>a</sup>	1.06±0.13 <sup>b</sup>	8.18*
Eicosa pentaenoic acid (g/100g)	0.43±0.08 <sup>a</sup>	0.35±0.02 <sup>a</sup>	0.50±0.03 <sup>a</sup>	2.45 (p = 0.17)
Docosa hexaenoic acid (g/100g)	0.32±0.09 <sup>a</sup>	1.05±0.05 <sup>a</sup>	1.04±0.48 <sup>a</sup>	3.53 (p = 0.10)
Total $\omega$ -3 (g/100g)	1.23±0.10 <sup>a</sup>	2.13±0.09 <sup>b</sup>	3.03±0.31 <sup>c</sup>	21.36*
Ratio $\omega$ -6 / $\omega$ -3	5.53±0.24 <sup>a</sup>	2.78±0.19 <sup>b</sup>	2.24±0.32 <sup>b</sup>	47.83*
AI index (g/100g)	6.71±0.48 <sup>a</sup>	4.15±0.29 <sup>b</sup>	3.75±0.11 <sup>b</sup>	23.62*
Milk yield (Kg/day)	37.48±1.18 <sup>a</sup>	22.79±2.55 <sup>b</sup>	19.22±0.98 <sup>b</sup>	29.58*

NG, non-grazing; HG, half-day grazing; FG, full-day grazing, <sup>a)</sup>Comparisons between the grazing time groups based on an analysis of variance (ANOVA), \*p < 0.05

From the above result, SFA, UFA, MUFA, EA, TN3, RN and AI were effective MFA to detect the optimum grazing time. Surely, these measurements had similar tendencies due to grazing time as we expected. The increase of grazing time resulted in the decrease of the SFA, RN and AI, while the increase of UFA, MUFA, EA and TN3. Two main milk fat acid factors were then generated for the grazing via principal component analysis ( $p < 0.05$ ), such as the positive milk fat acid (PMFA) associated with UFA, MUFA, EA and TN3 and the negative milk fat acid (NMFA) related to SFA, RN and AI.

### Determine the optimum grazing time

The optimum grazing time considering PMFA and MY was estimated by response surface methodology as described in Table 2. In detail, the UFA, MUFA, EA and TN3 (g/100g) were 17.82, 9.14, 0.61 and 2.06, respectively. The coefficient of determination was over 90 % ( $R^2 = 0.989$ ). Unfortunately, all parameters were not effective under the 5 % significance level. However, it was judged that the linear and interaction terms tend to effective except of quadratic term ( $p < 0.10$ ), considering the small sample size and number of level.

Table 2. The effects of positive milk fatty acid and milk yield on grazing time by response surface methodology (model  $R^2 = 0.989$ ).

Terms	Parameters	Estimate	Standard error	p-Value	R <sup>2</sup> (p-Value)
Linear	Intercept	-84.27	38.22	0.12	0.95*
	PMFA	74.86	25.25	0.06	
	Yield	7.27	2.84	0.08	
Quadra	PMFA x PMFA	-8.38	3.86	0.12	0.01 (p = 0.41)
	Yield x Yield	-0.14	0.05	0.08	
Interaction	PMFA x Yield	-2.41	0.92	0.08	0.03 (p = 0.08)

PMFA, positive milk fatty acid for grazing, \* $p < 0.05$

The optimum grazing time was hypothesized by tendencies of MFA and MY. There were two methods: one is checking a crossing point, other is estimating by RSM. The crossing point was 8.95 h between increase of PMFA and decrease of MY in shown as Figure 1 (A). When the optimum grazing time was recalling, the PMFA and MY were -0.19 and 25.46 kg/d, respectively. When the PMFA was recalling in detail, the UFA, MUFA, EA and TN3 (g/100g) were 17.68, 9.01, 0.59 and 2.03, respectively. The optimum grazing time by RSM was 11.05 h when the PMFA and MY were -0.08 and 27.16 kg/d respectively.

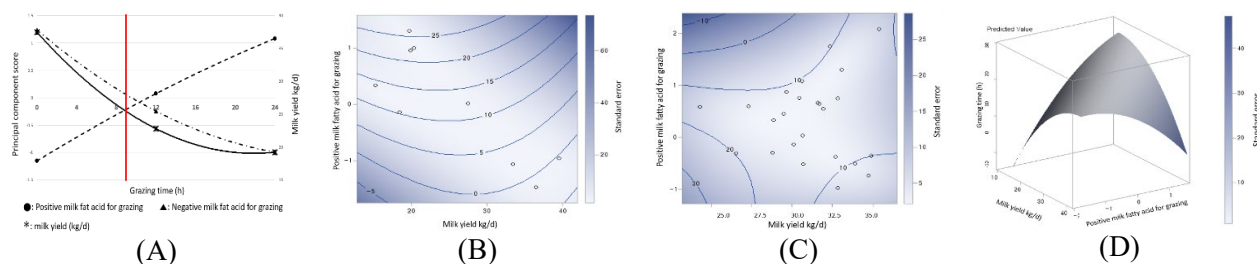


Figure 1. Line and contour plots of milk fatty acid and yield to select the optimum grazing time by response surface methodology: (A) line plot, (B) contour of surface, (C) contour of ridge, (D) contour of 3D surface.

Therefore, the tendencies of PMFA and MY were monotonously changed by increasing of grazing time. Furthermore, the single peak was obvious between increase PMFA and decrease MY (Figure 1B–D). In particular, white area related to the optimum time was stand out according to contour plot of ridge (Figure 1C).

In this study, assuming 100 % change of SFA and MUFA by grazing from 0 to 24 h, SFA and MUFA reached 58.68% decrease and 75.84 % increase respectively at the optimum grazing time. Furthermore the RN was decreased from 5.53:1 to 2.24:1 by grazing in alpine grassland. 150 years ago, the RN of human was originally 1:1 ratio, whereas, it has become in turns given rise to an imbalance in the RN by change in fatty acid consumption (Simopoulos, 2009). The proper ratio or balance between  $\omega$ -6 and  $\omega$ -3 fatty acids was important in the prevention and treatment of cardiovascular disease (Gómez Candela et al., 2011). Therefore, we judged that it was able to product good quality milk can be obtaining a premium grade for at least 11 hours of grazing in alpine grassland. Besides, economic analysis was required in order to evaluate the 27 kg/ha of MY at the optimum. The MY is crucial to farmers due to directly association to income. According to economic analysis for Holstein cows in this study, the MY decreased by 15 % (1028.21 USD/head), whereas, feed costs were

reduced by 43 % (399.54 USD/head) comparing with that of non-grazing system in the Republic of Korea (Sung et al., 2019). According to Balmford et al. (2008), reviews of wild nature loss was not about quantify the overall value of wild nature to human wellbeing like a loss of biodiversity and degradation due to infinity of nature. Furthermore, the MY could increase through suitable and continuous quality control like a pasture management, cow condition and barn management during the same grazing time (Hoogendoorn et al., 1992; Michel and Fulkerson, 1985). They reported that beneficial effect of increased grazing intensity on subsequent pasture quality and milk yields. Therefore, the optimum 11 hour of grazing time was likely to be minimum level, even considering not only direct and visible advantages, but also indirect and potential advantages from long-term environmental changes.

### **Discussion [Conclusions/Implications]**

About 11 hours grazing time was suggested focused on milk fatty acid and yield (27 kg/d) for Holstein cows as the optimum through eco-pastoral system. Although there was economic view based on prices of milk and forage, the optimum grazing time may be the minimum level since there was no prices can be taken into account the long termed and/or environmental benefits. This study will be helpful to select a time of grazing in alpine grassland for farmers who want to produce good quality milk.

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