1 Short running title: Goat grazing in abandoned fields

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- 3 Sustainable goat grazing for managing abandoned fields:
- 4 dynamics of vegetation quality, quantity and nutritional
- 5 status of goats over five years

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#### Abstract

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26Grazing goats have potential for use in managing abandoned 27fields. However, it is unclear how long-term grazing affects 28 goats as a result of vegetational changes in abandoned fields. 29This study aimed to assess the dynamics of vegetation quality 30 and quantity and the nutritional status of goats for five years 31 in an abandoned field at different stocking rates: high stocking rate (HS: 30-33 goats ha-1) and low stocking rate (LS: 3233 14 goats ha-1). In five years, the dominant plant species 34 changed from bamboo (Phyllostachys edulis) to tufted grass 35 species at the HS, whereas dwarf bamboo (Pleioblastus 36 argenteostriatus) dominated throughout the five years at the LS. 37 The annual plant biomass at both stocking rates was similar and varied from year to year. Crude protein content in the 38 39 goats' diet did not show any seasonal or annual trend regardless of the stocking rate. Neutral detergent fiber 40content in the HS diet did not show any seasonal or annual 41 trend; however, that in the LS diet linearly increased with the 42grazing year. The dry matter (DM) intake for goats at the LS 43 decreased linearly with the grazing year (P < 0.05), whereas 44 that for goats at the HS did not show any trend. The DM 45

46 digestibility was higher for goats at the HS than for goats at 47the LS (P < 0.05). Serum parameters did not show a negative nutritional status of the goats over the study years. Annual 48 49 daily gain was positive throughout the grazing years regardless of the stocking rate. Consequently, within the 50stocking rate range studied here, long-term goat grazing 5 1 52changes the quality of vegetation in an abandoned field; 53 however, goats can maintain their nutritional status and body weight for five years. Thus, goat grazing is a feasible way to 5455control abandoned vegetation over the long term.

- 57 Keywords
- 58 Abandoned field; goat grazing; nutrition; stocking rate;
- 59 sustainable management.

### Introduction

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62 The number of abandoned fields has been increasing over the 63 last four decades in developed countries (MacDonald et al. 642000; Cramer et al. 2008). In European countries and Japan, agricultural intensification and socioeconomic changes have 6566 generated many abandoned fields in marginal agricultural 67 areas (MacDonald et al. 2000; Katayama et al. 2015). Currently, the area of abandoned agricultural fields is over 6869 400,000 ha in Japan (MAFF 2017), and globally, the area of 70 abandoned fields is estimated to be from 385 to 472 million 7 1 ha (Campbell et al. 2008). 72In warm and wet climate areas, inadequate or poor field management leads to invasion and overgrowth of weedy plants, 73 and this often results in field degradation. Thus, an increase 7475in the area of abandoned fields induces the degradation of rural landscapes (MacDonald et al. 2000; Fukamachi et al. 76 77 2001) and the risk of wildfire (Osoro et al. 2007). Therefore, 78 appropriate, cost-effective and labor-saving management should be implemented to conserve these field environments. 79 80 Grazing could be a powerful tool for conducting this type of weed and field management. Goats may be effective in weed 81

82 management because they prefer shrubby and woody plants, 83 unlike cattle and sheep (Celaya et al. 2007a; Ferreira et al. 84 2013). Goats can survive in nutritionally harsh environments due to their ability to feed on a wide range of plants and their 85 86 low metabolic requirements (Lu 1988; Silanikove 2000). They are also suitable for hilly and mountainous environments in 87 88 marginal agricultural areas. Thus, some countries use goats 89 to control weedy plants (Popay and Field 1996; Celaya et al. 90 2010; Narvaez et al. 2012). 91 Generally, abandoned fields are composed of seminatural 92 vegetation that often contains fewer nutrients than sown 93 grasses, and their botanical composition differs among 94 regions and management regimens (Celaya et al. 2007b). For 95 example, European heathland communities consist of poorquality herbaceous plants (Celaya et al. 2007a; Osoro et al. 96 97 2007); thus, such field conditions limit the use of sustainable goat grazing systems (Osoro et al. 2013). In contrast, 98Japanese abandoned vegetation is composed of a variety of 99 plant species, such as grasses, sedges, legumes and forbs, and 100contains more than 50% total digestible nutrients (TDN) and 1018% crude protein (CP) (Tsutsumi et al. 2009). These values 102

would meet the recommendations of the NRC (2007) (more

- than 50% TDN and 7% CP) for the maintenance of goats. 104However, there is little information about whether such 105106 abandoned vegetation can support a sustainable goat grazing 107system over many years. 108 In grazing, the stocking rate is the primary factor that 109 affects vegetation dynamics (Celaya et al. 2010) and animal performance. Askar et al. (2013) revealed that a plant biomass 110 111 of more than 1,400 kg DM ha-1 did not restrict the 112performance of grazing goats regardless of stocking rate in a 113 grass/forb pasture. However, their study was carried out in a 114 sown pasture, and the results may not apply to abandoned 115fields because the quantity and quality of the vegetation in 116 abandoned fields are different from those in sown pastures. 117 Moreover, long-term grazing with different stocking rates 118 alters plant biomass and botanical composition (Marriott et
- Moreover, long-term grazing with different stocking rates alters plant biomass and botanical composition (Marriott et al. 2005; Celaya et al. 2010). Marriott et al. (2005) found a decrease in quantity and quality of forage with extensive grazing management over five years. Thus, vegetation changes that occur at an inappropriate stocking rate may hamper the use of sustainable grazing in the management of abandoned
- Therefore, understanding these dynamics will help in the

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fields

use of sustainable goat grazing systems to control abandoned agricultural land in a hilly area. The objective of the present study was to evaluate the quantitative and qualitative dynamics of vegetation, nutrient intake and goat performance in response to different stocking rates over a 5-year period and to determine a stocking rate at which goats are fed throughout the season only by grazing in abandoned fields.

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### Materials and methods

All animal experimental procedures were approved by the Committee for Animal Research and Welfare of Gifu University (#13022, 15020 and 17034) and were conducted in accordance with the guidelines of animal research and welfare of Gifu University.

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# Study site and experimental design

The experiment was conducted from 2013 to 2017 in an 142143abandoned field (0.8 ha) in Minokamo City, Gifu, central 144 Japan (35°29' N, 137°1' E, alt. 130 m). The average 145temperature and annual precipitation were 15.3 °C and 1841 146 mm, respectively, during the experimental years. The soil 147type in this region is brown forest soil based on volcanic 148 sediments. Approximately one-fourth of the study site was 149 composed of flat terrain, and the remaining area was hilly. 150The average gradient in the hilly part was 30 degrees. The site had been used for cultivation in the past, but after the 151 site was abandoned, invading plant species, mainly bamboo 152(Phyllostachys edulis), dominated the site for many years. 153Before the start of this experiment (March 2012), the bamboo 154155 was clear cut.

156The experimental site was divided into two areas (0.3 and 0.5 ha), and 16-17 Shiba×Saanen crossbreed goats (initial 157158mean body weight (BW):  $26.3 \pm 8.0$  kg in 2013,  $31.1 \pm 7.0$  kg in 2014,  $27.2 \pm 7.7$  kg in 2015,  $26.6 \pm 8.3$  kg in 2016 and 26.8159 160 $\pm$  9.3 kg in 2017) were assigned to the areas at two different 161 stocking rates: nine goats (2013-2016, 2017) or ten goats (5th year) represented a high stocking rate (HS: 30 or 33 goats ha-1621631), and seven goats represented a low stocking rate (LS: 14 164goats ha-1). The goats were stocked in the areas from May or 165June to October or November for five years (from 2013 to 166 2017). The grazing days were 146, 170, 165, 161 and 140 days 167in 2013, 2014, 2015, 2016 and 2017, respectively A wooden 168 shelter  $(3.6 \text{ m} \times 1.8 \text{ m} \times 1.8 \text{ m})$  was set before the 169 commencement of this study in each paddock. Animals had 170free access to mineral mixture and water tanks, and the water 171 was replaced every morning during the grazing period.

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#### Vegetation and animal measurements

Animal and vegetation measurements were conducted in the spring, summer and autumn, and each investigation period consisted of two weeks. In 2014-2017, the investigation dates were in spring (late May), summer (late July) and autumn

- 178 (mid-September-early October), but those in 2013 were one
  179 month later because the beginning of the goat grazing was
  180 delayed by one month.
- Botanical composition was measured at 20 fixed plots at each stocking rate during each investigation period using 25 cm × 25 cm quadrats. The plant species in the quadrats were identified, and classified into six plant groups: forbs, grasses, Cyperaceae, shrubs, bamboo and dwarf bamboo (*Pleioblastus argenteostriatus*). Then, the cover of each plant group in the quadrat was measured.
- 188 Plant biomass at each stocking rate was measured during 189each investigation period using 50 cm × 50 cm quadrats. Six 190 quadrats were randomly placed in each area, and plants within 191 a quadrat were cut at ground level and weighed. The plants 192were divided into edible and non-edible parts. Approximately 193200 g of the edible samples were collected, dried at 60 °C for 194 48 h and ground through a 1-mm screen using a Wiley mill. Dried samples were kept at room temperature until chemical 195196 analysis.
- Forage intake by the goats was measured using a doubleindicator method with n-alkane as an external indicator

  (Mayes et al. 1986) and acid detergent insoluble ash (ADIA)

200as an internal indicator (Nakano et al. 2007). n-Alkane capsules that included 0.25 mL of dotriacontane (C<sub>32</sub>) 201202solution were prepared. Each n-alkane capsule (C32) was 203administered with a small amount of concentrate to five 204mature goats in each stocking rate treatment at 9:00 am and 2054:00 pm every day during the investigation periods. Fecal grab samples were collected from the rectum of these goats 206207twice a day during the final week of each investigation period. Approximately 10 g of morning and evening fecal samples 208209 from each goat were well mixed and frozen at -30 °C and then 210 freezedried and milled through a 1-mm screen using a Wiley  $2 \ 1 \ 1$ mill. 212 To estimate dietary components and n-alkane contents of 213 the diets, we observed the foraging behavior of four goats 214that were used for intake measurements. The behavioral 215observations were conducted over two successive days during 216 each investigation period. On each day, two goats in each stocking rate treatment were observed by trained observers 217218for two hours during the morning and afternoon feeding bouts. 219The observers continuously counted the number of bites for each plant species. Then, the top 9-10 plant species in the 220bites (approximately 80-90% of the total number of bites) 221

collected by mimicking goats' feeding 222behavior. were Approximately 150 g of each plant species that was ingested 223224by the goats was imitatively collected by hand clipping. The plant samples were frozen at -30 °C and then freeze dried and 225226 milled through a 1-mm screen using a Wiley mill. The diet 227samples were prepared by mixing each ground plant sample according to the proportion of each plant in the bites. 228

Blood sampling was carried out at 10:00 am on the final day of each period. Blood samples (20 mL) were collected from the jugular vein of the five goats that were used for intake measurements. Blood samples were immediately placed on ice and centrifuged at  $1,000\times g$  at 4 °C for 20 min. Serum was separated and frozen at -30 °C until analysis.

The goats were weighed on the first and last days of each investigation period.

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### Chemical analyses

Plant biomass, diet and fecal samples were analyzed for dry matter (DM), CP and crude ash (CA) content according to the Association of Official Analytical Chemists procedures (AOAC 2007). Neutral detergent fiber (aNDFom) was assayed with heat-resistant amylase and ash, acid detergent fiber was

expressed exclusive from residual ash (ADFom) and ADIA 244245contents were determined according to Van Soest et al. (1991). 246The contents of n-alkane  $(C_{32})$  in fecal samples were measured according to the method of Dove and Mayes (2006) 247248using gas chromatography (GC) with a flame ionization 249detector. The GC sample analysis used a capillary column (TC Series TC-1, 30 mm × 250 µm diameter, GL Sciences, Tokyo, 250251Japan). The GC system was set at an injector and detector temperature of 350 °C. The GC oven program included an 252253initial temperature of 200 °C, which was held for 0.5 min, 254 and according to the program, the temperature then changed by 20  $^{\circ}\text{C}$  min  $^{\text{-1}}$  to 250  $^{\circ}\text{C}$  , 10  $^{\circ}\text{C}$  min  $^{\text{-1}}$  to 300  $^{\circ}\text{C}$  , 6  $^{\circ}\text{C}$  min  $^{\text{-1}}$ 255256to 324 °C and 3 °C min-1 to 350 °C, and the system was then 257held at this final temperature for 2 min. Serum glucose, total cholesterol (TCHO), urea nitrogen 258259(UN), total calcium (Ca), magnesium (Mg) and inorganic 260phosphate (IP) contents were analyzed using the Dry-Chemistry system (DRI-CHEM 4000V, Fujifilm Medical, 261262Tokyo, Japan) in the first year of the study and using a Discrete Automated Clinical Chemistry Analyzer (Accute 263264TBA-40FR, Canon Medical Systems, Tochigi, Japan) in the 265other years.

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- 267 Estimation of fecal output, plant digestibility and dietary
- 268 intake
- 269 Fecal output, plant digestibility and daily dietary intake were
- 270 estimated using the following equations (Dove and Mayes
- 271 2006):
- 272 Fecal output (kg DM day<sup>-1</sup>) =  $C_{32}$  dose rate (mg day<sup>-1</sup>)/ $C_{32}$
- 273 fecal concentration (mg kg DM<sup>-1</sup>)
- 274 Digestibility (%) = (1 (ADIA g kg DM<sup>-1</sup> in diet)/(ADIA g kg
- $275 \quad DM^{-1} \text{ in feces}) \times 100$
- 276 DM intake (kg DM day<sup>-1</sup>) = (fecal output [kg DM day<sup>-1</sup>]/(1 -
- 277 digestibility)
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- 279 Statistical analysis
- 280 Plant intake and digestibility, daily gain (DG) and serum
- 281 blood components were analyzed using a generalized linear
- 282 mixed model. Fixed effects were stocking rate, season, year
- 283 and their interactions. Individual variation among goats was
- 284 considered as a random effect. The statistical procedures were
- 285 performed using the ImerTest package (Kuznetsova et al.
- 286 2014) in R software (ver. 3.0.2.) (R Core Team 2013). If the
- 287 effect of season and year were significant (P < 0.05),

288 orthogonal contrasts were performed.

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290 Results

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### Botanical composition and plant biomass

293 Bare ground and litter gradually disappeared with progression of the grazing years in both treatments (Table 1). 294295The cover of the forb group was the highest in both treatments 296 throughout the five years. Similarly, the cover of grasses in 297 the LS treatment modestly increased, but dwarf bamboo 298 maintained its cover at approximately 15-20% during the five 299years. 300 The annual plant biomass at both stocking rates was similar 301 for five years (Table 2), although the biomass at 302 beginning of the experiment (spring in 2013) was different 303 between the two treatments (HS: 4.9 t DM ha-1; LS: 3.5 t DM 304ha-1). Plant biomass decreased with the progress of seasons at both stocking rates. In brief, the average plant biomass in 305spring was approximately 3 t DM ha-1 (HS: 3.3 and LS: 2.8 t 306 DM ha-1), but biomass fell below 2 t DM ha-1 in autumn (HS: 3071.8 and LS: 1.6 t DM ha-1). Plant biomass varied from year to 308

year. In the HS treatment, the biomass in the first year showed

the highest value, decreased in the second year and then gradually increased in the remaining years. In the LS treatment, the biomass was highest in the first year and dropped below 2.0 t DM ha<sup>-1</sup> in the other years, except for the fourth year.

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# Diet compositions

The goats in both treatments mainly bit the forbs during the first year of grazing, but this tendency gradually decreased with the progress of the grazing years (Table 3). In HS goats, percentage of bites of grasses and bamboos increased over five years, whereas in LS goats, that of bamboo and dwarf bamboo increased over the period.

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### Chemical compositions of diets

The CP content in the HS diet decreased from spring to summer (14.3 to 13.2%) and then increased again in autumn (15.0%) (Table 4). However, the CP content in the LS diet linearly increased from spring to autumn (12.6 to 13.6%). The aNDFom content in the HS diet increased from spring to summer (45.1 to 54.5%) and decreased in autumn (51.6%), whereas that of the LS diet increased from spring to autumn

- 332 (54.1 to 57.0%). The ADIA content of diets in both treatments
- 333 linearly increased with the progression of seasons (HS: 2.4
- 334 to 6.5% and LS: 3.3 to 8.8%).
- 335 The CP content in the HS diet was higher than in the LS
- 336 diet (14.2 and 13.1%, respectively), and the aNDFom and
- 337 ADIA contents were higher in the LS diet (aNDFom: 55.4 and
- 338 ADIA: 5.8%) than in the HS diet (aNDFom: 50.4 and ADIA:
- 339 4.7%).
- 340 The CP content did not show any annual trend and varied
- 341  $\,$  from 12.4 to 15.8% in the HS diet and from 12.4 to 14.1% in
- 342 the LS diet. The aNDFom content in the HS diet also showed
- 343 no annual trend (44.0 to 55.6%), whereas that in the LS diet
- 344 linearly increased from the first to fifth year (51.1 to 60.8%).
- 345 The ADIA content in the HS diet was between 4.3 and 5.1%
- 346 and was almost constant over the five years. In the LS diet,
- 347 the ADIA content showed an increasing trend from the first
- 348 to the fifth year (4.8 to 6.2%).
- 350 Daily gain

- 351 The average DG of the grazing goats did not differ between
- 352 the stocking rate treatments; DG was 7.0 g day-1 in the HS
- 353 treatment and 16.6 g day-1 in the LS treatment (Table 5).

Season significantly and quadratically (P < 0.05) affected DG at both stocking rates; that is, DG was lower in summer (HS: -39.4 and LS: 12.8 g day<sup>-1</sup>) than in other seasons (spring, HS: 58.4 and LS: 67.4 g day<sup>-1</sup>; autumn, HS: 65.8 and LS: 44.3 g day<sup>-1</sup>). However, annual DG was positive regardless of the stocking rate and was not significantly affected by year (Table 5).

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#### Intake

363 Stocking rate did not affect DM, CP or aNDFom intake (Table 364 5). In contrast, DM intake was significantly affected by 365season and by the stocking rate × season interaction (season: 366 P < 0.05, stocking rate × season: P < 0.05). The average DM intake for goats at the HS was 83.8 g/kg BW0.75 in spring but 367368 decreased with season (Figure 1, linear response: P < 0.05), 369 while the goats at the LS showed higher DM intake in summer than in spring and autumn (Figure 1, quadratic response: P <370 0.05). Season also affected aNDFom intake (quadratic 371 response: P < 0.05). The aNDFom intake at both stocking rates 372ranged from 36.7 to 44.6 g/kg  $BW^{0.75}$  and was higher in 373 summer than in spring and autumn (quadratic response: P <3740.05). Season did not affect CP intake at either stocking rate, 375

but the stocking rate  $\times$  season interaction significantly affected CP intake (P < 0.05). Specifically, CP intake for goats at the HS was relatively constant throughout the grazing season, whereas their CP intake at the LS showed higher values in summer than in spring and autumn (Figure 1, quadratic response: P < 0.05).

The DM, CP and aNDFom intake at both stocking rates was

383 significantly different among grazing years (P < 0.05). The DM 384intake at both stocking rates was highest in the second year 385 and lowest in the last year (Figure 2). For goats at the LS, 386 DM intake decreased with year (Figure 2, linear response: P 387< 0.05), while that of goats at the HS did not show any trend. 388 The CP intake at both stocking rates varied from 6.9 to 13.6 389 g/kg BW<sup>0.75</sup> (Table 5) among the years. The CP intake for 390 goats at the LS was highest in the second year and lowest in 391 the fifth year (Figure 2. quadratic response, P < 0.05). For 392goats at the HS, CP intake was almost constant throughout grazing years. The aNDFom intake for goats at the HS showed 393 a quadratic response with grazing years (Figure 2, P < 0.05), 394 while aNDFom intake for goats at the LS decreased linearly 395396 with grazing years (P < 0.05).

# Digestibility

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The DM and aNDFom digestibility significantly differed 399 400between the stocking rates (P < 0.05). The DM digestibility 401was higher in goats at the HS (53.8%) than in goats at the LS 402(45.3%), and aNDFom digestibility was also higher in goats at the HS (49.7%) than in goats at the LS (43.4%). Stocking 403404rate did not show any significant effect on CP intake. 405Season significantly affected DM, C P a n d a N D F o m 406digestibility (DM: P < 0.05, CP: P < 0.05, aNDFom: P < 0.05) 407(Table 5). DM digestibility in goats at the HS decreased from 408 61.4% in spring to 46.7% in autumn (linear response: P <0.05) (Figure 1), whereas goats in the LS treatment showed 409410 higher DM digestibility in summer (52.0%) than in spring and 411 autumn (49.0 and 33.4%; quadratic response: P < 0.05). 412Seasonal changes in CP digestibility showed a trend similar 413 to that of DM digestibility at both stocking rates, but there was no significant seasonal effect on CP digestibility in goats 414 415at the HS (Figure 1). The aNDFom digestibility was the highest in summer (HS: 52.8 and LS: 50.7%) and the lowest 416 in autumn (HS: 45.8 and LS: 35.1%) at both stocking rates 417418 (Figure 1). The aNDFom digestibility in goats at the LS showed a quadratic response (P < 0.05) (Figure 1), while no 419

420 significant trend was detected in goats at the HS.

421There was no effect of year on DM digestibility, but year 422significantly affected CP and aNDF om digestibility (CP: P <0.05, aNDFom: P < 0.05) (Table 5). aNDFom digestibility 423424showed a quadratic response regardless of stocking rate (quadratic response: P < 0.05) (Figure 3). There was a 425significant difference in CP digestibility among the years, 426427with no linear or quadratic changes detected at both stocking 428rates (Table 5 and Figure 2). Annual aNDFom digestibility at 429both stocking rates showed a quadratic response according to 430 year (quadratic response: HS, P < 0.05; LS, P < 0.05) (Table 5 431and Figure 2).

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# Serum blood components

- 434 Stocking rate did not affect blood components except for Mg
- 435 (Table 5). The Mg concentration was higher in goats at the
- 436 HS (3.0 mg  $dL^{-1}$ ) than in goats at the LS (2.8 mg  $dL^{-1}$ ).
- 437 A seasonal effect was found on serum glucose, TCHO, UN
- 438 and Mg concentrations (all: P < 0.05); however, there were
- 439 no significant stocking rate × season interactions for any
- 440 serum components. Serum glucose concentrations at both
- 441 stocking rates were lower in summer than in spring and

autumn (quadratic response: P < 0.05) (Figure 3). Although 442differences 443there significant i n serum TCHO were concentrations among seasons, differences were not as great 444445throughout the gazing seasons (Figure 3). Serum 446 concentrations in the grazing goats increased linearly from 44717.0 to 22.9 mg dL<sup>-1</sup> regardless of the stocking rate (Figure 3). Serum Mg concentrations increased from 2.8 to 3.0 mg dL 448449 <sup>1</sup> with the progression of seasons in goats at the HS (P < 0.05)450(Figure 3), whereas that in goats at the LS did not show any significant change with season. Season did not affect serum 451452Ca and IP concentrations at either stocking rate (Ca: 9.1 to 4539.2 mg dL<sup>-1</sup> at the HS and LS; IP: 5.5 to 6.0 mg dL<sup>-1</sup> at the 454HS and 6.5 to 6.7 mg  $dL^{-1}$  at the LS) (Table 5 and Figure 3). 455With respect to the effect of years, a significant difference 456was found in serum glucose, UN, Ca and Mg concentrations 457(all: P < 0.05) (Table 5), and a significant stocking rate  $\times$ 458year interaction was detected for serum glucose concentration (P < 0.05). Serum glucose concentrations in the goats at the 459460HS were relatively constant over the five-year period, whereas that in the goats at the LS increased over the grazing 461462years (P < 0.05) (Figure 4). Serum UN concentrations decreased linearly in the LS treatment (P < 0.05) (Figure 4), 463

464and the UN concentration was 22.6 mg dL-1 in the first year but was  $19.2~\text{mg}~\text{dL}^{-1}$  in the fifth year. Serum TCHO 465466 concentrations did not show any significant change with years (ranging from 83.9 to 89.4 mg dL<sup>-1</sup>) (Figure 4). Serum Ca 467468concentrations s h o w e d quadratic response a a n d M g 469 concentrations showed a linear increase with grazing year (Figure 4). Ca concentrations showed the lowest value in the 470471third year, and Mg concentrations increased from 2.9 to 3.1 mg dL-1 over the five years. Year did not affect serum IP 472473 concentrations at either stocking rate (HS: 5.4 to 6.3 mg dL-474 <sup>1</sup>, LS: 6.2 to 7.0 mg dL<sup>-1</sup> over the five-year period).

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# Discussion

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#### Stocking rate

The annual stocking rates were 828 and 386 kg ha<sup>-1</sup> at the HS and LS, respectively. Stocking rate did not affect DM, CP or aNDFom intake or the DG of the goats (Table 5). Generally, stocking rate influences plant biomass in a pasture and forage intake by goats (Animut and Goetsch 2008). In the present study, the plant biomass at both stocking rates was maintained at more than 1,600 kg DM ha<sup>-1</sup> throughout the grazing seasons.

486The CP content was higher than 12% and the aNDFom content was less than 56% in the diets regardless of season and year, 487488 suggesting that diets contained moderate nutritive value. 489 Askar et al. (2013) reported that stocking rate did not limit 490 the DG of grazing goats when they grazed on a pasture with 491 1,400 kg DM ha-1 or more of plant biomass and a moderate nutritive value. The present results agree with those previous 492493 results, and the presence of sufficient plant biomass at both 494stocking rates resulted in no restriction on the DG of the 495goats. 496 Both DM and aNDFom digestibility were influenced by the 497stocking rate. These differences were caused by differences 498 in the diet compositions between stocking rate treatments 499 (Table 3). The grasses and bamboo (Phyllostachys edulis) in 500the diet of HS goats increased over five years, while the 501bamboo and dwarf bamboo in the diet of LS goats increased 502throughout the experiment (Table 3). The percentage of bites of shrubs, which are often high in NDF and lignin contents 503504(Papachristou and Nastis 1993), in LS goats was also higher 505than that in HS goats. The differences in diet composition

Specifically, the aNDFom and ADFom contents in the diet

might relate to the chemical composition of the goat's diet.

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508were greater for goats at the LS (55.4 and 31.9%) than the HS (50.4 and 29.9%). Similarly, ADIA contents in the diets were 509slightly but unalterably higher for goats at the LS than for 510goats at the HS. Accordingly, DM and fiber digestibility in 511 512 goats at the LS were lower than in those at the HS. Thus, the 513 original differences in botanical composition between areas different stocking rates induced differences 514t h e 515chemical composition in the diets and resulted in the different 516 digestibilities.

517 Serum Mg concentration was affected by the stocking rate 518 (P < 0.05). Both paddocks included a variety of plant species, 519and generally, Mg content differed in each plant species 520(Yoshihara al. 2013). Thus, different botanical e t 521m a y have contributed t o different Μg 522concentrations in both groups; however, grazing goats at both 523stocking rates did not show any symptoms of Mg deficiency 524during the study.

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# Effects of grazing season

The DM intake for goats at the HS ranged from 66.5 to 83.8 528 g/kg BW<sup>0.75</sup> and decreased linearly from spring to autumn. In 529 contrast, intake for the goats at the LS ranged from 68.1 to

- 530 g/kg BW<sup>0.75</sup> and changed from spring to autumn. According to the NRC feeding standards, mature goats at a 531 BW of 30 kg require 53 g/kg BW<sup>0.75</sup> of DM intake for 532 maintenance (NRC 2007). Thus, grazing goats at both stocking 533 rates satisfied their maintenance requirements throughout the 534 535 grazing seasons. The DM digestibility decreased linearly in goats at the HS 536 537 and showed an opposite trend in goats at the LS (P < 0.05), 538although digestibility at both stocking rates was the lowest
- Plant maturation involves 539 i n autumn. a n increase 540indigestible components due to lignification of cell walls 541(Kozloski et al. 2005). Accordingly, plant digestibility in 542ruminants decreases with increasing plant maturity. In the 543 present study, the ADIA content of the goats' diet at both 544 stocking rates showed the highest value in autumn. Thus, diet 545digestibility at both stocking rates had the lowest value in 546autumn.
- Serum glucose and TCHO concentrations are indices of the energy status of ruminants (Ndlovu et al. 2007). In the present study, serum TCHO concentrations in grazing goats were relatively constant during grazing seasons regardless of the stocking rate. Conversely, glucose concentrations

decreased in summer at both stocking rates. Ndlovu et al. 552(2007) reported that heat stress increased body temperature 553 and respiratory rate and resulted in a decrease in blood 554 glucose concentrations in cattle. Several studies have also 555 556 observed a decrease in glucose concentrations in grazing cattle in summer (Grünwaldt et al. 2005; Mapiye et al. 2010). 557The results of the present study are consistent with those of 558 559 the above previous studies. Thus, the decrease in serum 560glucose concentrations in summer was probably caused by 561 heat stress rather than by insufficient energy intake. 562 Serum UN concentrations increased from spring to autumn 563 at both stocking rates. The concentrations were influenced by 564 the CP/energy ratio in the diet (Hammond et al. 1994; 565Karnezos et al. 1994). Increasing serum UN concentrations in 566 grazing goats suggest that CP and energy ratios in the diet were imbalanced with the progress of seasons, even in the 567 568 vegetation of abandoned fields. 569 Serum mineral concentrations at both stocking rates were almost constant throughout the grazing seasons, except for 570571 serum Mg concentrations in goats at the HS (Figure 3). A simulation study by Yoshihara et al. (2013) suggested that 572

increasing the number of plant species reduces the risk of

both excesses and deficiencies in ingested minerals. In the 574 present study, goats ate more than 20 plant species at both 575 576 stocking rates. Thus, grazing on abandoned fields, which are 577 composed of many plant species, may contribute to stabilizing 578 serum mineral concentrations in goats.

579 Grazing goats at both stocking rates showed the lowest, and negative, values in average DG in summer, although intake 580 and digestibility in summer were higher than in autumn. 582Generally, heat stress increases the energy consumption of 583 grazing goats by increasing the respiration rate (Shinde et al. 584 2002), suggesting that forage species in abandoned fields 585cannot provide sufficient energy to overcome the increase in 586 energy requirements induced by heat stress. However, such 587 negative impact on nutritional status was only observed in 588 summer, and the annual DG of grazing goats at both stocking 589 rates did not show negative values. These results suggest that 590the vegetation in abandoned fields in central Japan can support the maintenance requirements of grazing goats 591throughout the grazing season. 592

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# Effects of grazing year

Forage biomass at both stocking rates did not show

596 decreasing trend with the progress of the grazing year, and its annual variation might be due to annual meteorological 597598 conditions. The chemical composition of the diets differed 599 between the stocking rates. In the HS treatment, the values 600in the diet were relatively constant for five years, whereas 601the quality of the diet, which was reflected by CP and aNDFom 602contents, decreased over the 5-year period in the LS treatment. 603This continuing decrease in diet quality might induce a linear 604decrease in the DM intake of goats at the LS. However, DG in 605both groups did not decrease over the five years, suggesting 606 that the vegetation in the abandoned fields can support the 607maintenance requirements of grazing goats for five years 608 regardless of the stocking rate. 609 During five years, s e r u m glucose a n d TCHO 610 concentrations in grazing goats at both stocking rates did not 611 fall below the hypoglycemic and hypocholesterolemic values 612in goats (37.8 and 65.8 mg dL<sup>-1</sup>, respectively; Žubčić 2001). Serum glucose concentrations linearly increased with the 613 614 grazing year, and the reason for this increase was not clear. 615Serum UN concentrations showed a decreasing trend with the 616 grazing year (P < 0.05), although a significant effect was not detected in the HS treatment. However, serum UN levels were 617

close to 20 mg dL<sup>-1</sup>, which was the upper limit of standard values (NLBC 2018), suggesting that the energy/protein ratio in the diet was not desirable even in the vegetation of abandoned fields, as is the case in sown grasses.

622Serum Ca, IP and Mg concentrations at both stocking rates 623were relatively stable during the five years; however, serum Ca and Mg concentrations differed among the years. Moreover, 624625all the serum mineral concentrations were within the range of 626standard values for goats (Kaneko et al. 1997). Mineral 627concentrations are highly variable among plant species 628(Ohlson and Staaland 2001), and increasing the number of 629plant species reduces the risk of excesses and deficiencies of 630 ingested minerals (Yoshihara et al 2013). In the present study, 631 grazing goats ingested more than 40 plant species each year. 632Consequently, abandoned fields that contain various plant 633 species can provide diets that are suitable for maintaining 634 stable serum mineral concentrations in grazing goats over a 635period of five years.

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#### Conclusion

638 Goat grazing over five years changed the botanical 639 composition in an abandoned field. This change in vegetation

640affected the chemical composition of the diet but did not affect plant biomass over five years. Although DM intake at 641642the LS linearly decreased with grazing year, the overall DG 643 remained positive throughout the grazing years regardless of 644the stocking rate. Serum parameters at both stocking rates did 645not indicate a negative nutritional status over the years studied, except for relatively higher UN concentrations. 646647Serum mineral concentrations were relatively stable because 648grazing goats ingested various plant species in the abandoned 649field. Thus, within a stocking rate range (14-33 goats ha-1), 650the use of grazing for the sustainable management of 651abandoned fields is feasible and does not have negative 652effects on the nutritional status of goats.

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# Conflict of interest statement

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Table 1 Changes in botanical compositions with season and year at different stocking rates

	2013			2014			2015			2016			2017		
Cover (%)†	Spring	Summer	Autumn												
High stocking rate															
Bareground and litter	23.8	44.6	43.3	41.3	40.4	38.0	7.4	20.8	23.5	17.8	21.1	14.1	14.8	19.4	15.6
Forbs	43.8	14.6	24.7	36.2	34.5	43.4	57.1	41.7	48.6	48.7	31.6	42.1	51.2	31.3	39.3
Bamboo	18.8	22.7	14.0	1.7	7.0	3.0	3.7	12.2	3.8	7.9	15.6	9.9	6.9	16.5	11.9
Dwarf bamboo	0.0‡	1.2	0.8	5.0	0.7	1.6	1.3	1.2	1.0	1.0	2.5	2.3	4.9	4.9	3.1
Grasses	4.9	7.0	11.5	10.9	12.3	12.5	26.4	19.5	17.5	16.0	20.8	21.3	18.2	24.1	23.0
Cyperaceae	7.0	8.0	5.0	0.0	4.9	0.8	2.1	2.8	3.5	6.0	6.8	6.3	3.9	2.3	5.6
Shrubs	1.8	1.8	0.8	4.8	0.3	0.8	2.1	1.8	2.1	2.6	1.7	4.0	0.1	1.5	1.5
Low stocking rate															
Bareground and litter	34.1	51.4	51.2	40.1	42.3	42.8	26.0	26.1	24.0	14.5	17.0	18.3	10.3	17.9	22.1
Forbs	29.5	11.0	11.8	35.1	16.4	10.5	32.7	24.8	28.3	32.9	26.9	28.3	32.9	26.9	35.8
Bamboo	7.5	11.5	10.7	0.0	9.0	7.4	6.4	8.7	3.4	7.4	10.8	10.6	2.3	9.6	8.1
Dwarf bamboo	17.0	15.2	11.9	14.5	13.5	16.8	12.9	18.9	15.2	16.7	18.1	13.9	21.5	19.1	18.9
Grasses	2.5	0.8	10.3	6.4	9.8	12.7	17.7	15.7	19.3	20.7	19.5	13.3	16.4	11.7	11.4
Cyperaceae	2.0	5.0	2.0	0.8	5.8	7.3	3.7	4.0	8.0	4.7	4.4	3.7	6.6	9.4	7.5
Shrubs	7.3	5.2	2.0	3.1	3.3	2.5	0.7	1.7	1.8	3.2	3.2	4.4	3.6	3.2	2.2

<sup>†</sup>Means of all investigation plots (20 points in each treatment).

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<sup>†</sup>Bamboo is *Phyllostachys edulis*, and Dwarf bamboo is *Pleioblastus argenteostriatus*.

<sup>‡</sup>This value is less than 0.1%.

Table 2 Changes in plant biomass (DM) with season and year at different stocking rates

Item	SR†	Season			Year				
		Spring	Summer	Autumn	2013	2014	2015	2016	2017
Biomass (t DM ha <sup>-1</sup> )	HS	3.3	1.9	1.8	3.0	1.9	2.1	2.3	2.5
	LS	2.8	2.0	1.6	2.6	1.8	1.8	2.5	2.0

†HS: 30 goats per ha; LS: 14 goats per ha.

Table 3 Changes in diet compositions (percentage of bites) of grazing goats with season and year at different stocking rates

	2013			2014		·	2015			2016			2017		
Diet composit	ion Spring	Summer	Autumn	Spring	Summer	Autumn									
(%)†															
High stocking rate	:														
Forbs	82.5	45.9	43.8	29.7	26.9	23.2	59.2	27.6	6.5	54.2	26.5	30.9	59.6	18.6	15.8
Bamboo	0.1	5.5	25.2	15.9	37.6	19.3	1.5	5.2	14.5	13.4	33.9	24.8	5.7	36.6	33.5
Dwarf bamboo	10.5	1.9	2.0	29.2	15.5	5.0	14.2	13.2	22.4	2.6	1.9	4.6	4.6	9.9	2.0
Grasses	2.3	28.6	17.1	13.1	11.7	36.2	17.1	44.3	52.3	12.4	23.5	24.1	20.8	14.0	31.2
Cyperaceae	2.3	10.6	9.4	8.3	7.8	14.8	7.0	6.8	3.9	4.1	11.9	11.7	5.8	18.1	16.5
Shrubs	2.3	7.5	2.4	3.9	0.5	1.5	1.0	2.9	0.3	13.3	2.2	3.6	3.5	2.8	0.7
Litter	0.0‡	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2	0.0	0.0	0.4	0.0	0.0	0.3
Low stocking rate															
Forbs	58.2	34.8	14.1	37.3	30.0	19.4	30.1	24.0	21.3	32.8	31.2	20.7	20.0	12.2	9.8
Bamboo	1.0	1.5	4.0	27.6	10.2	19.4	27.9	26.8	40.0	14.7	19.3	28.5	47.0	35.0	29.3
Dwarf bamboo	17.1	2.3	28.3	8.2	0.5	22.0	5.1	9.0	0.6	32.3	22.5	21.4	13.1	29.5	29.4
Grasses	16.6	34.9	27.1	7.7	32.5	11.1	14.4	27.2	20.9	8.3	14.2	14.0	11.1	18.6	19.0
Cyperaceae	1.9	6.9	8.4	3.5	17.7	17.8	8.3	9.6	12.7	9.3	9.8	9.6	1.6	3.4	10.1
Shrubs	5.2	19.5	16.9	15.3	8.7	7.7	13.9	3.3	2.2	2.2	3.0	5.2	6.7	1.3	1.9
Litter	0.0	0.1	2.1	0.4	0.3	2.6	0.3	0.0	2.2	0.4	0.0	0.5	0.5	0.0	0.5

<sup>†</sup>Bamboo is *Phyllostachys edulis*, and Dwarf bamboo is *Pleioblastus argenteostriatus*.

<sup>‡</sup>This value is less than 0.1%.

Table 4 Changes in chemical composition in goat's diets with season and year at different stocking rates

Item†	Overall mean	Season			Year							
,		Spring	Summer	Autumn	2013	2014	2015	2016	2017			
HS												
DM (%)	27.5	27.9	26.3	28.2	26.2	27.0	29.8	27.1	27.5			
CP (%)	14.2	14.3	13.2	15.0	15.0	12.4	15.8	14.3	13.3			
aNDFom (%)	50.4	45.1	54.5	51.6	44.0	55.6	47.3	53.8	51.1			
ADFom (%)	29.9	29.1	31.4	29.3	28.2	31.7	27.0	31.8	30.9			
ADIA (%)	4.7	2.4	5.3	6.5	4.5	4.9	4.9	4.3	5.1			
CA (%)	11.4	8.9	12.0	13.3	11.8	11.3	11.4	10.9	11.5			
LS												
DM (%)	27.5	26.3	25.4	30.7	26.9	26.4	29.7	26.0	28.2			
CP (%)	13.1	12.6	13.2	13.6	14.1	13.6	12.6	12.9	12.4			
aNDFom (%)	55.4	54.1	55.1	57.0	51.1	53.7	54.7	56.7	60.8			
ADFom (%)	31.9	31.2	32.5	31.8	31.8	31.0	30.7	32.7	33.1			
ADIA (%)	5.8	3.3	5.3	8.8	4.8	4.9	7.0	5.9	6.2			
CA (%)	12.2	9.5	12.5	14.6	12.3	11.9	12.4	12.2	12.2			

†DM: Dry matter; CP: Crude protein; aNDFom: Neutral detergent fiber assayed with hieat-stable amylase and expressed exclusively of residual ash; ADFom: Acid detergent fiber expressed exclusively of residual ash; ADIA: Acid detergent insoluble ash; CA: Crude ash.

**Table 5** Changes in daily gain, dietary intake and plant digestibility of each nutrient category and serum blood component in grazing goats at different stocking rates and in different seasons and years

Item	SR†		Season			Year					Significanc	e level			
	HS	LS	Spring	Summer	Autumn	2013	2014	2015	2016	2017	SR	Season	Year	0.007 <0.001 0.154 <0.001 0.001 0.054 0.238 0.634	$SR \times Ye$
Daily Gain (g day-1)	7.0	16.6	64.4	-17.0	54.6	10.6	13.7	16.5	12.0	5.8	0.179	< 0.001	0.764	0.177	0.147
Intake (g/kg BW <sup>0.75</sup> )															
DM	76.8	73.8	75.1	82.3	68.1	73.9	102.6	67.5	80.6	53.1	0.197	0.002	< 0.001	0.007	0.304
CP	10.9	9.7	10.1	11.0	9.9	10.8	13.6	9.5	11.1	6.9	0.081	0.061	< 0.001	< 0.001	0.019
aNDFom	38.8	40.4	36.9	44.6	36.7	35.1	55.7	34.1	44.1	28.8	0.515	< 0.001	< 0.001	0.154	0.521
Digestibility (%)															
DM	53.8	45.3	55.9	52.2	40.7	46.0	51.2	50.0	53.6	48.9	0.007	< 0.001	0.208	< 0.001	0.113
CP	59.0	51.3	57.4	56.6	52.1	51.9	52.1	55.9	60.8	56.9	0.172	0.011	0.007	0.001	0.011
aNDFom	49.7	43.4	47.0	51.8	41.0	33.5	48.9	52.1	52.3	47.3	0.003	< 0.001	< 0.001	0.054	0.048
Serum blood component (mg dL <sup>-1</sup> )															
Glucose	49.0	51.4	50.9	44.5	54.9	50.6	45.2	51.8	46.9	55.4	0.273	< 0.001	< 0.001	0.238	0.028
Urea Nitrogen	20.5	20.8	17.0	22.2	22.9	22.6	22.1	19.1	20.4	19.2	0.882	< 0.001	0.042	0.634	0.893
Total cholesterol	86.9	88.3	84.1	90.7	88.0	89.3	90.5	85.3	89.4	83.9	0.488	0.023	0.446	0.767	0.204
Ca	9.2	9.1	9.2	9.1	9.2	9.5	9.4	8.6	9.1	9.1	0.812	0.352	< 0.001	0.564	0.814
IP	5.8	6.6	6.2	6.2	6.1	6.4	6.1	6.3	6.1	6.2	0.364	0.922	0.505	0.556	0.428
Mg	3.0	2.8	2.8	2.8	3.0	2.9	2.4	2.8	3.1	3.1	0.048	< 0.001	< 0.001	0.668	0.202

†HS: 30 goats per ha; LS: 14 goats per ha.

- 864 Figure legends
- 865
- 866 Figure 1 Seasonal changes in DM, CP and aNDFom intake and
- 867 digestibility in grazing goats (means  $\pm$  SE for five-year
- 868 results). HS indicates 30-33 goats per ha, and LS indicates
- 869 14 goats per ha. In the statistical analysis, L, Q and NS
- 870 indicate the results of an orthogonal contrast analysis. L
- 871 represents a seasonally linear trend, Q represents a
- 872 seasonally quadratic trend, and NS indicates no trend.
- 873
- 874 Figure 2 Changes in DM, CP and aNDFom intake and
- 875 digestibility in grazing goats with year (means ± SE for five-
- 876 year results). HS indicates 30-33 goats per ha, and LS
- 877 indicates 14 goats per ha. In the statistical analysis, L, Q and
- NS indicate the result of an orthogonal contrast analysis. L
- 879 represents a linear trend, Q represents a quadratic trend, and
- NS represents no trend in relation to the year.
- 881
- 882 Figure 3 Seasonal changes in serum glucose, TCHO, UN, Ca,
- 883 IP and Mg concentrations in grazing goats (means  $\pm$  SE for
- 884 five-year results). HS indicates 30-33 goats per ha, and LS
- 885 indicates 14 goats per ha. In the statistical analysis, L, Q and

NS indicate the results of an orthogonal contrast analysis. L<br/>
represents a seasonally linear trend, Q represents a<br/>
seasonally quadratic trend, and NS indicates no trend.

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Figure 4 Changes in serum glucose, TCHO, UN, Ca, IP and Mg concentrations in grazing goats according to year (means ± SE for five-year results). HS indicates 30-33 goats per ha, and LS indicates 14 goats per ha. In the statistical analysis, L, Q and NS indicate the results of an orthogonal contrast analysis. L represents a linear trend, Q represents a quadratic trend, and NS represents no trend according to year.







