

Silvopasture versus open-pasture systems for broilers

**Effect of silvopasture system on fearfulness and leg health in fast-growing
broiler chickens**

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21 **Abstract**

22 A silvopasture system intentionally integrates trees, forages, and livestock, allowing dual
23 land use. These systems can provide high-quality habitat for broiler chickens; however, such
24 systems have not been widely adopted by the broiler industry in the United States. The objective
25 of this study was to examine the effect of silvopasture versus open pasture access on fearfulness
26 and leg health in fast-growing broiler chickens. A total of 886 mixed-sex Ross 708 chicks in
27 Experiment 1 (Exp 1) and 648 chicks in Experiment 2 (Exp 2) were housed in coops and had
28 access to 16 (Exp 1) or 12 (Exp 2) 125m² silvopasture plots (\bar{x} = 32% canopy cover) or open
29 pasture plots (no canopy cover) from day 24 of age. Fearfulness was measured using a tonic
30 immobility test (tonic immobility duration), and leg health was assessed by quantifying footpad
31 dermatitis, hock burns, gait, and performing a latency-to-lie test on days 37-39 of age. Birds in the
32 silvopasture treatment were less fearful than birds in the open pasture treatment. Overall, birds in
33 both silvopasture and open pasture systems had excellent leg health. Silvopasture birds had lower
34 footpad dermatitis scores than open pasture birds. Silvopasture birds tended to have worse gait
35 than open pasture birds in Exp 1, but not in Exp 2. Hock burn scores and latency-to-lie did not
36 differ between treatments in Exp 1 or Exp 2. Raising birds in silvopasture reduced fear and
37 improved footpad health compared to birds raised in open pastures, which indicates that
38 silvopasture systems provide some benefits for affective state and leg health in fast-growing
39 broilers.

40

41 **Keywords:** Agroforestry, animal welfare, broiler chicken, fearfulness, leg health, outdoor
42 access

43

44 **Introduction**

45 The United States (US) has three main broiler chicken production systems which mandate
46 that birds have outdoor access. The first is certified organic production overseen by the United
47 States Department of Agriculture (USDA), which requires that birds have access to the outdoors,
48 with shade, shelter, exercise areas, fresh air, direct sunlight, and protection from predators year-
49 round [1]. The second is free-range poultry production, overseen by the USDA, where producers
50 must meet the required housing conditions to be able to sell products using the free-range label
51 [2]. These conditions include continuous free access to the outdoors for more than 51% of the
52 animals' lives [2]. The third production system is pastured poultry production, which could be
53 certified by a number of voluntary assurance schemes. Depending on those assurance schemes
54 requirements for outdoor access can vary. Some animal-welfare benefits of these systems
55 compared to indoor-only housing systems include more space and more opportunities to perform
56 natural behaviors [3–6].

57 Broiler chicken production with outdoor access requires more land than indoor-only
58 systems. Furthermore, birds may have negative experiences accessing the pasture when faced with
59 extreme temperature fluctuations, presence or threat of predators, and exposure to diseases and
60 parasites, compared to indoor-only conditions [7–9]. These conditions can increase production
61 costs and may lead to decreased gain compared with birds in indoor housing. For instance, lower
62 final body weights and higher feed conversion ratios were found for broilers with outdoor access
63 compared to indoor only fast-growing birds [10]. To compensate for the additional land costs as
64 well as potential impacts on animal performance, land productivity can be increased by utilizing a
65 silvopasture system. A silvopasture system is an actively managed tree-forage-livestock system on
66 a single piece of land [11,12]. Short-term income can be generated through the production of

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67 poultry products, and long-term income can be obtained through tree or forage production. In
68 addition, fruits and browse can have value for consumption by humans or broilers, which further
69 helps to improve short-term economic returns by reducing food or feed costs [11]. Along with
70 these economic benefits, silvopasture systems can reduce environmental stressors and serve as
71 high-quality habitat for broiler chickens compared to open pasture. Yet, silvopasture systems have
72 not been widely adopted by the broiler chicken industry.

73 In current production systems that allow outdoor access, ranges commonly consist of open
74 grassland habitats [6]. Grass pastures typically do not provide birds opportunities to seek cover
75 from aerial predators [10,13]. Chickens prefer trees and shrubs because they provide natural cover
76 to hide [14]. Access to pasture with trees or hedgerows can prevent predation loss, provide a milder
77 microclimate, and improve productivity and physiological health. Olive trees or sorghum in the
78 range resulted in no or <1% mortality due to predation for male naked-neck chickens, compared
79 to 2.4-2.8% mortality in flocks kept in open grass pastures [15]. The microclimate in a mature-
80 pine silvopasture system was milder compared to an open pasture system nearby in Florida, US
81 [16]. Similarly, mean wet-bulb globe temperatures were lower in silvopasture than open pastures
82 on the same farm in Virginia, US [17]. Access to a pasture with a mature hedgerow compared to
83 an open pasture resulted in improved weight gain without increasing feed costs in fast-growing
84 broilers [18].

85 Access to a pasture with trees rather than no or artificial shelters could potentially benefit
86 birds' emotional reactivity, such as fearfulness. Stadig and colleagues [19] found that far-ranging
87 slow-growing broilers were less fearful with access to willows than close-ranging broilers that had
88 access to artificial shelters. The complex environment of a silvopasture may reduce fearfulness
89 similarly as indoor-only environments with enrichment objects [20,21]. For instance, broilers or

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90 broiler breeders were less fearful when housed with perches [22], music [23], dust baths and
91 temporary objects [21] such as balls, strings, or wall drawings [24,25] compared to broilers without
92 enrichments. The potential benefit for reducing fearfulness in fast-growing broilers with
93 silvopasture access compared to open pastures has not yet been studied, even though these fast-
94 growing broilers are the common strains used in certified-organic and some free-range and
95 pastured poultry production systems in the US.

96 In conventional indoor-only broiler chicken production systems, animal welfare concerns
97 include poor leg and foot health. In a heavy-bird (~3.7 kg) indoor-only production system in the
98 US, 47.5% of birds had superficial or deep footpad lesions [26]. These lesions, referred to as
99 footpad dermatitis (FPD), are problematic from an economic [27,28] and animal welfare
100 perspective [29]. Broilers can experience pain associated with FPD [30] and will be less motivated
101 to access feeders and drinkers. These animals gain less weight than broilers without FPD and thus
102 have lower carcass yields [29,31,32]. Providing broilers with outdoor access can reduce the
103 prevalence of FPD. The outdoor space reduces in-house stocking density, increasing birds' space,
104 and thereby reducing point contamination with feces and moisture within the house. For instance,
105 higher litter moisture levels were found in a barn system (38% vs. 28%) compared to a free-range
106 system for turkeys between 18 and 20 weeks of age [33]. The lower litter moisture concentrations
107 indoors reduce the chances of developing contact dermatitis, including FPD, breast dermatitis and
108 hock burn lesions [29,32,34,35]. Access to an outdoor range would therefore reduce chances of
109 FPD and hock burn lesion development in two ways, (1) by distributing fecal material across the
110 range (thus decreasing fecal material deposition in house) and (2) by reducing birds' time sitting
111 inside, which could lessen their contact with moisture and feces. A silvopasture system may
112 encourage even greater range utilization compared to an open pasture in fast-growing broilers. For

113 example, range use in medium-growing broilers was improved with access to conifer wigwams
114 compared to birds in open pasture [36]. Similarly, range use in slow-growing broilers with access
115 to short rotation coppice was 7% greater compared to birds with artificial shelters in the range [37].
116 In commercial operations (indoor-only, heavy-bird production) in the US, 34.5% of birds had
117 abnormal gait or were totally lame [26]. Over 27% of indoor-only housed broilers had poor
118 walking ability in the United Kingdom, with 3% of birds having limited ability to walk at 40 days
119 of age [38]. Outdoor access could improve gait and reduce lameness. Slow-growing broilers that
120 used their outdoor range had improved gait compared to birds that were indoors [39] or used the
121 range less [19]. Yet, the potential benefit of silvopasture access for improving leg health in fast-
122 growing broilers has not been studied.

123 Several studies have shown animal welfare benefits of outdoor access for fast-growing
124 broilers [18,40], slow-growing broilers [19,39,41,42], and turkeys [33]. However, it is unclear
125 whether the welfare benefits of pasture access, and especially silvopasture access, can be obtained
126 for fast-growing broiler chickens, which are the most common type of broiler chicken produced in
127 the US. Therefore, our study objective was to evaluate the effect of silvopasture versus open
128 pasture access on fearfulness and leg health in fast-growing broiler chickens. We hypothesized
129 that birds in silvopasture systems would be less fearful and have improved leg health (FPD, hock,
130 gait, and latency to lie) than birds in open pasture systems.

131 **Materials and Methods**

132 **Animals and housing**

133 Two experiments were conducted from April to May (Experiment 1; Exp 1) and June to
134 August 2021 (Experiment 2; Exp 2). All procedures were approved by the Virginia Tech
135 Institutional Animal Care and Use Committee (IACUC protocol 20-044).

136 In total, 886 one-day-old Ross 708 mixed-sex chicks in Exp 1 and 648 chicks in Exp 2
137 were obtained from a commercial hatchery (Harrisonburg, VA, USA). Birds were Marek's
138 vaccinated at the hatchery. Upon arrival, chicks were arbitrarily selected and housed in 12 identical
139 pens (5.7 m²) with 73 or 74 birds per pen in Exp 1 and 53 or 54 birds per pen in Exp 2. Pens
140 contained pine wood shavings (~5 cm depth), a heat lamp (day 1-8), a cardboard feed flat with
141 feed (day 1-8), one bell drinker (Plasson® Broiler Drinker complete, Or-Akiva, Israel), and one
142 pet champion poultry drinker (Stout Stuff LLC, China), and one feeder (Superbowl poultry feeder,
143 LaGrange, NC, USA). The chicks were fed commercial starter (day 0-15), grower (day 15-25),
144 and finisher (day 25-42 or 43) diets meeting requirements [43]. Ambient temperatures were 35°C
145 on day 1 and gradually reduced to 23°C on day 22 (Exp 1) or day 23 (Exp 2). Lighting was
146 provided continuously for the first week and reduced to 12h light and 12h dark until day 22 (Exp
147 1) or day 23 (Exp 2).

148 In Exp 1, birds from each pen were equally and randomly allocated over 16 pasture-based
149 treatments resulting in 53 birds per plot. In Exp 2, complete pens (53-54 birds) were randomly
150 allocated to 12 pasture-based plots with chicken coops. On day 22 (Exp 1) or day 23 (Exp 2), birds
151 were transported for 1.5h to the pasture-based experimental site located at the Shenandoah Valley
152 Agricultural Research and Extension Center (AREC) in Raphine, VA, US. After transportation,
153 birds were kept inside the coops for two days (Exp 1) or one day (Exp 2) to get acclimated to their

154 new housing conditions. From day 24, coop doors in each plot were opened at approximately 8:00
 155 AM and closed at approximately 5:00 PM.

156 All pasture-based plots (125m²) contained a chicken coop (6.55m²) constructed from wood,
 157 chicken wire, and tarp [44]. Each coop contained a wooden platform perch (0.05m × 0.10m ×
 158 2.40m), feeder, and bell drinker. Coops were moved laterally across the plot each week. Plots were
 159 fenced with 1-m-high and 50-m-long FlexNet electric fences (PoultryNet®, Washington, IA,
 160 USA), connected to a 30-volt electric cattle fence. Mean pen and coop stocking densities on day
 161 1, day 22 or 23, and day 42 or 43 for Exp 1 or Exp 2 are given in Table 1.

162

163 **Table 1. Mean (± SEM) of pen and coop stocking density on day 1, day 22-23, and day 42-43**
 164 **of age in Experiments 1 (Exp 1) and 2 (Exp 2).**

Stocking Density	Pen		Coop			
	Day 1		Day 22		Day 42	
	g/m ²	Birds/m ²	kg/m ²	Birds/m ²	kg/m ²	Birds/m ²
Exp 1	518.1±0.8	13.0±0.0	7.5±0.1	8.0±0.0	20.8±0.4	7.2±0.1
	Day 1		Day 23		Day 43	
Exp 2	439.3±13.3	9.4±0.0	7.4±0.1	8.2±0.0	21.8±0.2	8.1±0.1

165

166 **Treatments**

167 The silvopasture plots (four replicates in two locations in Exp 1, and three replicates in two
 168 locations in Exp 2; Fig 1) contained a mixed hardwood stand of black walnut (*Juglans nigrea* L.),
 169 locust (*Robinia pseudoacacia* L.) and hickory (*Carya* spp. Nutt.) trees and 30 newly planted

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170 saplings per plot (American hazelnut (*Corylus americana*), black walnut (*Juglans nigra*),
171 persimmon (*Diospyros virginiana* L.), southern red oak (*Quercus falcata* Michx.), and southern
172 pine (*Pinus* spp.) of approximately 30-cm height and 1-cm diameter. Saplings were planted in six
173 rows with inter- and intra-row spacing of 1.5m. Canopy cover in all silvopasture and open pasture
174 plots was calculated from photos using ImageJ software (1.5.3k, National Institutes of Health,
175 Bethesda, MD, USA). The images were taken straight upwards from ground level in the center of
176 the plot. Photos were converted to 8-bit, binarized, and then the number of black (canopy) and
177 white (sky) pixels were calculated as a percentage of total pixels. The canopy cover for silvopasture
178 plots was (mean±standard deviation) 31.7±16.7% in Exp 1, and 33.3±10.9% in Exp 2. Open
179 pasture plots had no canopy cover.

180 The open pasture plots (four replicates in two locations in Exp 1 and three replicates in two
181 locations in Exp 2; Fig 1) contained ground vegetation. Common herbaceous forage species in the
182 open pasture and silvopasture plots were tall fescue (*Schedonorus arundinaceus* (Schreb.)
183 Dumont., syn. *Lolium arundinaceum* (Schreb.) Darbysh., formerly *Festuca arundinacea* Schreb.),
184 orchard grass (*Dactylis glomerata* L.), horse nettle (*Solanum carolinense* L.), and common
185 milkweed (*Asclepias syriaca* L.). Forage species specific to silvopasture plots includes greenbrier
186 (*Smilax* spp. L.), honeysuckle (*Lonicera japonica* Thunb.), Virginia creepers (*Parthenocissus*
187 *quinquefolia* L. Planch.), nimble will (*Muhlenbergia schreberi* J.F. Gmel.), poison ivy
188 (*Toxicodendron radicans* (L.) Kuntze ssp. *radicans*), wild basil (*Clinopodium vulgare* L.),
189 common blue violet (*Viola sororia* Willd), and wood sorrel (*Oxalis* spp.). Forage species specific
190 to open pasture plots were common chickweed (*Stellaria media* (L.) Vill.), common dandelion
191 (*Taraxacum officinale* F.H. Wigg.), and Queen Anne's lace (*Daucus carota* L.).

192

193 **Fig 1. Image and illustration of experimental plots (125m² each).** Open grassland pasture plots
194 (orange) and silvopasture plots (black/white) contained a chicken coop (black). Experiment 1
195 included 4 replicates per treatment in two locations, and Experiment 2 included 3 replicates per
196 treatment in two locations. The omitted replicate plots for Experiment 2 are marked with a pattern
197 fill. Map sourced from USGS National Map Viewer.

198

199 **Measurements**

200 A total of 79 or 80 birds per treatment (159 birds total) in Exp 1 and 78 birds per treatment
201 (156 birds total) in Exp 2 received a numbered leg band on day 37 or 38 of age for individual
202 identification. In Exp 1, 10 birds per plot were sampled; in Exp 2, 13 birds per plot were sampled.
203 All measurements were performed on the same birds. As sampling took place on pasture, observers
204 could not be blinded to experimental treatments.

205

206 **Tonic immobility**

207 Birds were tested for tonic immobility (TI) duration and TI induction on day 38 in Exp 1
208 and day 37 of age in Exp 2. Two observers performed the TI test in both experiments. The inter-
209 observer agreement was tested for 10 birds and was excellent (Cronbach's α of 0.92). The TI test
210 was conducted as described by [21]. However, birds were tested outdoors in the pasture rather than
211 indoors. During the TI test, the assessed birds did not have visual contact with other birds or the
212 observer. A bird was placed on its back in a V-shaped cradle, and then TI was induced by
213 restraining the bird with one hand on its sternum for 15 seconds while covering their head with the
214 other hand. At the end of the induction period, both hands were gently removed. If the bird tried
215 to right themselves within 10 seconds, the induction attempt was considered failed, and the handler

216 repeated the restraint procedure (no more than three induction attempts). In Exp 1, one bird for
217 which TI could not be induced was replaced with another bird. In Exp 2, birds that could not be
218 induced were included in the sample and received a TI duration of 0 seconds. After successful
219 induction of TI, the TI duration was recorded for a maximum of 5 minutes.

220

221 **Footpad dermatitis and hock burn lesions**

222 Eighty birds per treatment (160 birds total) in Exp 1 and 77 or 78 birds per treatment (155
223 birds total) in Exp 2 were assessed for FPD and hock burn lesions on day 39 in Exp 1 and day 38
224 in Exp 2. FPD and hock burn lesions were scored on a 0-4 categorical scale, with increasing scores
225 indicating worse lesions [45]. A single trained observer scored FPD and hock burn lesions in Exp
226 1 and Exp 2 and recorded the most severe score of a bird's two feet or hocks.

227

228 **Gait**

229 Birds were evaluated individually for their walking ability and assigned a categorical gait
230 score between 0-2, with higher scores representing worse gait [46]. A single trained observer
231 performed the scoring by voluntarily allowing the birds to walk for at least 1.5m. If the birds did
232 not walk, the observer stimulated the bird by gently touching their tail or vent with a rod. If the
233 bird did not walk after gentle stimulation, the bird received the highest gait score (score 2).

234

235 **Latency to Lie**

236 Fifty-five or 56 birds per treatment (111 birds total) in Exp 1 and 77 birds per treatment
237 (154 birds total) in Exp 2 were individually assessed in a latency to lie (LTL) test on days 39 or
238 40 in Exp 1 and days 37 or 38 in Exp 2. The test was performed as described by [47]. The LTL

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239 was recorded as an indicator of leg strength, with shorter latencies representative of poorer leg
240 strength [47]. Individual birds were placed in an opaque plastic tub (0.93m L × 0.54m W × 0.46m
241 H; Sterilite Corporation, Townsend, MA, USA) containing 2-3cm lukewarm (29 - 36°C) water for
242 a maximum of 10 minutes. In Exp 2, the tub was covered with a barrier made from bird netting
243 and PVC pipe to avoid escape behaviors observed in Exp 1. Four birds were tested simultaneously
244 in separate tubs. Birds that remained standing at the end of the test received an LTL of 10 minutes.
245 A single observer performed the tests.

246

247 **Weather and soil conditions**

248 Ambient temperature (°C), photosynthetically active radiation ($\mu\text{Em}^{-2}\text{s}^{-1}$), soil moisture (%
249 volumetric moisture content), relative humidity (%), and dew point (°C) were measured using one
250 Spectrum WatchDog 1000 Series MicroStation (Spectrum Technologies, Inc., Aurora, IL, USA)
251 per treatment from day 27 to day 43 in Exp 1.

252

253 **Statistical analysis**

254 Data of both experiments were analyzed separately in JMP Pro 16 (SAS Institute Inc., Cary,
255 NC, USA). Data residuals of continuous response variables were assessed for the normality of
256 their distribution by visual inspection of normal quantile plots. Data residuals of TI duration and
257 LTL were not normally distributed. TI duration was log-transformed and analyzed using a mixed
258 model with treatment as fixed factor and plot as random factor. TI attempts (counts) were analyzed
259 in SAS Studio 3.8 (SAS Institute Inc., Cary, NC, USA) with a linear mixed model with a Poisson
260 distribution, using treatment as fixed factor and plot as random factor. Transformation of LTL did
261 not result in normality of residuals; thus, plot means were calculated to obtain a single measure

262 per experimental unit and avoid pseudo-replication. These were analyzed in a mixed model with
263 treatment as fixed factor and plot as random variable. Ordinal response variables (FPD, hock, and
264 gait scores) were analyzed in SAS Studio using generalized linear mixed models with a
265 multinomial (ordered) distribution, with treatment as fixed factor and plot as random factor.
266 Outliers were not removed from the dataset. The threshold for significance was set at $p \leq 0.05$ and
267 for a trend at $p \leq 0.10$. Raw means and standard errors are reported unless otherwise noted.

268

269 **Results**

270 **Tonic immobility**

271 Silvopasture birds tended to show shorter TI durations than open pasture birds in Exp 1
272 ($F_{1,14}=3.89$, $p=0.069$) and were shorter in Exp 2 ($F_{1,10}=24.04$, $p<0.001$; Fig 2). No difference was
273 found in the number of attempts to induce TI among birds in the silvopasture and open pasture
274 treatments in Exp 1 ($F_{1,143}=0.19$; $p=0.664$) or Exp 2 ($F_{1,143}=0.73$; $p=0.395$).

275

276 **Fig 2. Means for tonic immobility (TI) duration (\pm SEM) for broilers raised in silvopasture**
277 **or open pasture plots on day 38 in Experiment 1 (n=159) and on day 37 of age in Experiment**
278 **2 (n=156).** ^{AB} Raw means within Experiment 1 with uncommon superscripts differ at $p<0.10$. ^{ab}

279 Raw means within Experiment 2 with uncommon superscripts differ at $p<0.05$.

280

281 **Leg health**

282 Silvopasture birds tended to have lower (improved) FPD scores than open pasture birds in
283 Exp 1 ($F_{1,141}=3.30$; $p=0.071$) and had lower scores in Exp 2 ($F_{1,143}=5.61$; $p=0.019$; Fig 3). Mean

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284 FPD scores were 0.08 ± 0.04 (Exp 1) and 0.17 ± 0.05 (Exp 2) for silvopasture birds, and 0.19 ± 0.05
285 (Exp 1) and 0.48 ± 0.08 (Exp 2) for open pasture birds. Hock burn scores did not differ between
286 treatments in Exp 1 ($F_{1,142}=2.14$; $p=0.146$) or Exp 2 ($F_{1,143}=2.66$; $p=0.105$; Fig 4). Mean hock burn
287 scores were 0.15 ± 0.05 (Exp 1) and 0.03 ± 0.02 (Exp 2) for birds in the silvopasture treatment, and
288 0.26 ± 0.05 (Exp 1) and 0.09 ± 0.03 (Exp 2) for birds in the open pasture treatment. In Exp 1 but not
289 in Exp 2, silvopasture birds tended to have worse gait scores than open pasture birds ($F_{1,143}=2.96$;
290 $p=0.088$ in Exp 1 and $F_{1,143}=1.44$; $p=0.231$ in Exp 2). The majority of sampled birds showed
291 excellent gait, with $\geq 80\%$ of birds having a gait score 0 (Fig 5). Mean gait scores were 0.21 ± 0.05
292 (Exp 1) and 0.05 ± 0.03 (Exp 2) for silvopasture birds, and 0.08 ± 0.03 (Exp 1) and 0.10 ± 0.04 (Exp
293 2) for open pasture birds. No difference was found in LTL between silvopasture birds and open
294 pasture birds in Exp 1 ($F_{1,14}=1.04$, $p=0.324$) and Exp 2 ($F_{1,10}=1.22$, $p=0.295$). The mean LTL was
295 452 ± 28 sec (Exp 1) and 523 ± 19 sec (Exp 2) for birds in silvopasture, and 403 ± 28 sec (Exp 1) and
296 549 ± 13 sec (Exp 2) for birds in open pasture.

297

298 **Fig 3. Percentage of birds with footpad dermatitis (FPD) scores 0-4 in the silvopasture and**
299 **open pasture treatments.** (a) On day 39 of age in Experiment 1 (n=159). (b) On day 38 of age in
300 Experiment 2 (n=155).

301

302 **Fig 4. Percentage of birds with hock burn scores 0-4 in the silvopasture and open pasture**
303 **treatments.** (a) On day 39 of age in Experiment 1 (n=159). (b) On day 38 of age in Experiment 2
304 (n=155).

305

306 **Fig 5. Percentage of birds with gait score 0-2 in the silvopasture and open pasture treatments.**

307 (a) On day 39 of age in Experiment 1 (n=160). (b) On day 38 of age in Experiment 2 (n=155).

308

309 **Weather and soil conditions**

310 Ambient temperature, photosynthetically active radiation, soil moisture, relative humidity,

311 and dew point in the silvopasture and open pasture treatments in Exp 1 are presented in Table 2.

312 Although not statistically analyzed, all but one (soil moisture) values were lower in the silvopasture

313 treatment compared to the open pasture treatment.

314

315 **Table 2. Raw means (\pm SEM) of weather and soil conditions in the silvopasture and open**

316 **pasture treatments from day 27 to day 43 in Experiment 1 (n=2).**

Weather and soil conditions	Silvopasture	Open pasture
Ambient temperature ($^{\circ}\text{C}$)	14.1 \pm 0.1	15.0 \pm 0.2
Photosynthetically active radiation ($\mu\text{Em}^{-2}\text{s}^{-1}$)	171.4 \pm 4.6	272.4 \pm 6.7
Soil moisture (% volumetric moisture content)	99.8 \pm 0.1	16.8 \pm 0.2
Relative humidity (%)	64.8 \pm 0.3	70.9 \pm 0.2
Dew point ($^{\circ}\text{C}$)	6.5 \pm 0.1	8.0 \pm 0.1

317

318 **Discussion**

319 This study investigated the effect of silvopasture versus open pasture access on fearfulness

320 using a TI test and on leg health using FPD scores, hock burn scores, gait scores, and LTL in fast-

321 growing broiler chickens. We found that pasture type impacted fearfulness, FPD, and tended to

322 impact gait scores, with silvopasture access showing improvements for all measures besides gait,
323 and no difference in hock burns or LTL.

324

325 **Tonic immobility**

326 The shorter TI duration in birds in the silvopasture treatment in Exp 1 and Exp 2 indicates
327 that those birds were less fearful than birds in the open pasture treatment [48]. TI durations in the
328 current study were shorter than durations reported in previous research with conventional broilers
329 raised in different housing conditions; indoor-only [10,42,49,50], indoor housing with outdoor
330 access [10,49–51], and indoor housing with environmental enrichment [10,24,52–54]. TI durations
331 (103-104 sec) in our open pasture treatments were comparable to the duration of a slow-growing
332 broiler strain housed with outdoor access (108 sec; [42]. Far-ranging slow-growing broilers were
333 less fearful with access to willows than close-ranging broilers with access to artificial shelters [19],
334 suggesting that trees in the range were more meaningful to reduce fearfulness than artificial
335 shelters in their study. This is in line with findings in our study, as we also found birds with access
336 to trees to be less fearful than birds without trees in the range. Lack of overhead cover in open
337 pasture plots could have increased the birds' chance of encountering predators, increasing broilers'
338 fear and anxiety [55] compared to birds in the silvopasture plots. In the current study no mortality
339 from areal predation occurred, yet one account of predation by a ground predator was observed in
340 an open pasture plot (Exp 1), thought to be caused by a raccoon (*Procyon lotor*) when birds were
341 42 days of age, which was 4 days after fear was assessed.

342 In addition to being obscured from predators, we argue that a more complex outdoor range
343 with trees could serve a similar function as enrichments provided when housed indoors, either
344 biologically relevant (perches) or biologically less relevant (such as music or balls), as both

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345 enrichments and access to a complex range promote the expression of natural behaviors
346 [3,5,56,57]. Broilers raised with environmental complexity in indoor-only housing conditions were
347 less fearful compared to broilers housed in simpler conditions. For instance, Arbor Acres birds
348 with music [23] and Ross 708 birds with dust baths, perches, and temporary objects [21] were less
349 fearful than without enrichments. Our results indicate that allowing fast-growing broiler chickens
350 access to a silvopasture range reduces fearfulness compared to access to an open pasture range.

351

352 **Footpad dermatitis and hock burn lesions**

353 Broilers with access to silvopasture had better FPD scores than broilers with access to open
354 pasture. Five percent of birds in Exp 1 and 15% of birds in Exp 2 had mild or severe FPD scores
355 in the silvopasture treatment, which is lower or comparable to reported prevalences of 12% and
356 22% in slow-growing broilers with access to olive trees or sorghum [41], and lower than
357 prevalences of 68% and 72% in slow-growing broilers with access to willows or artificial shelters
358 [19]. Litter (soil) moisture content is identified as a key risk factor for the development of contact
359 dermatitis [32,35,58]. The lower FPD prevalence and severity in silvopasture plots, even with
360 higher soil moisture content (Table 2), may be due to increased range use in silvopasture plots
361 compared to open pasture plots, which could have reduced the in-coop stocking density, reduced
362 fecal material within the coop, and reduced birds' time sitting inside. Despite the soil moisture
363 content being higher in the silvopasture treatment compared to the open pasture treatment, our
364 findings show that FPD prevalence and severity in fast-growing broilers are minor in either pasture
365 system, but especially minimal in the silvopasture system. Although not statistically assessed, FPD
366 scores were lower in Exp 1 compared to Exp 2. Lower maximum in-coop stocking density in Exp
367 1 versus 2 (Table 1) could have contributed to that difference. Seasonal variation in FPD scores in

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368 fast-growing indoor-only broilers was reported, with the best outcomes during the warmest months
369 [59], yet those results do not align with our findings, as FPD was slightly worse in summer
370 compared to spring.

371 Hock health was excellent in birds with access to either open pasture or silvopasture and
372 did not differ between treatments. Similar to FPD scores, severity and prevalence of hock burns
373 were low in both treatments. Although no previous work has investigated hock burn lesions in
374 fast-growing broilers with pasture access, our findings generally do align with earlier research on
375 slow-growing broilers with outdoor access [40,42]. Hock burn scores for silvopasture and open
376 pasture birds in the current study were better when scored on a 5-point scale (mean scores between
377 0.03-0.26) than reported for slow-growing broilers from seven organic farms with outdoor access
378 scored using a similar 4-point scale (mean score of 0.30; [42]). Birds in the silvopasture treatment
379 had comparable or higher hock burn scores as slow-growing broilers from 25 farms with outdoor
380 access (score 0=96% and score 1=4%; [40]), while birds in the open pasture treatment showed
381 more severe hock burn lesions compared to those reported by [40]. These results met our
382 expectations as FPD and hock burn lesions can show a positive association and share some of the
383 same etiology [60].

384

385 **Gait**

386 Most birds in the current study had excellent gait, with the majority showing no gait
387 imperfection in either treatment group. Outdoor access has been associated with better gait in fast-
388 growing [51,61] and slow-growing broilers [19,51] compared to indoor-only housing. Although
389 [10] found no difference in gait scores in fast-growing broilers raised indoors compared to broilers
390 with outdoor access. Even though access to pasture resulted in good gait across treatments, gait

391 tended to be worse in the silvopasture treatment compared to the open pasture treatment in Exp 1
392 only. This is contrary to our hypothesis and does not align with the outcomes for other leg health
393 indicators that were assessed, where silvopasture resulted in an improvement or no difference. The
394 mechanism behind the difference in gait scores in Exp 1 is unclear. The lack of a difference in Exp
395 2 suggest that it may not be consistently due to the pasture treatment, but may have been a result
396 of other factors.

397 **Latency to Lie**

398 The LTL test measures leg strength in broilers by recording the time it takes for birds to lie
399 down in lukewarm water [62], relying on chickens' aversion to sit down in water, therefore, shorter
400 latencies reflect poorer leg strength [47]. LTL did not differ between treatments in either
401 experiment. In the current study, birds showed comparable LTL, thus suggesting comparable leg
402 strength to slow-growing strains (mean LTL between 403-548 sec vs. 547 sec reported by [42].
403 Generally, leg health is worse in fast-growing broilers compared to slow-growing strains [63]. This
404 suggests that both treatments in the current study resulted in good leg strength in fast-growing
405 broilers, which is a major achievement in terms of animal welfare, as leg issues are a well-
406 recognized welfare concern for fast-growing broilers [38,64].

407

408 **Conclusion**

409 This study evaluated the effect of silvopasture versus open pasture systems on fearfulness
410 and leg health in fast-growing broiler chickens. To our knowledge, this is the first study to assess
411 the impact of a pasture system (silvopasture or open pasture) on aspects of animal welfare and
412 health of fast-growing broiler chickens. We found that providing fast-growing broilers with access

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413 to a silvopasture system from 3 weeks of age is particularly beneficial for reducing fearfulness and
414 improving leg health, specifically footpad condition, compared to providing access to open
415 pasture. Even though gait was worse in silvopasture-raised broilers in one of two experiments,
416 scores were low (thus gait was not impaired) in either treatment. Leg strength was good and
417 comparable in both treatments.

418

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423

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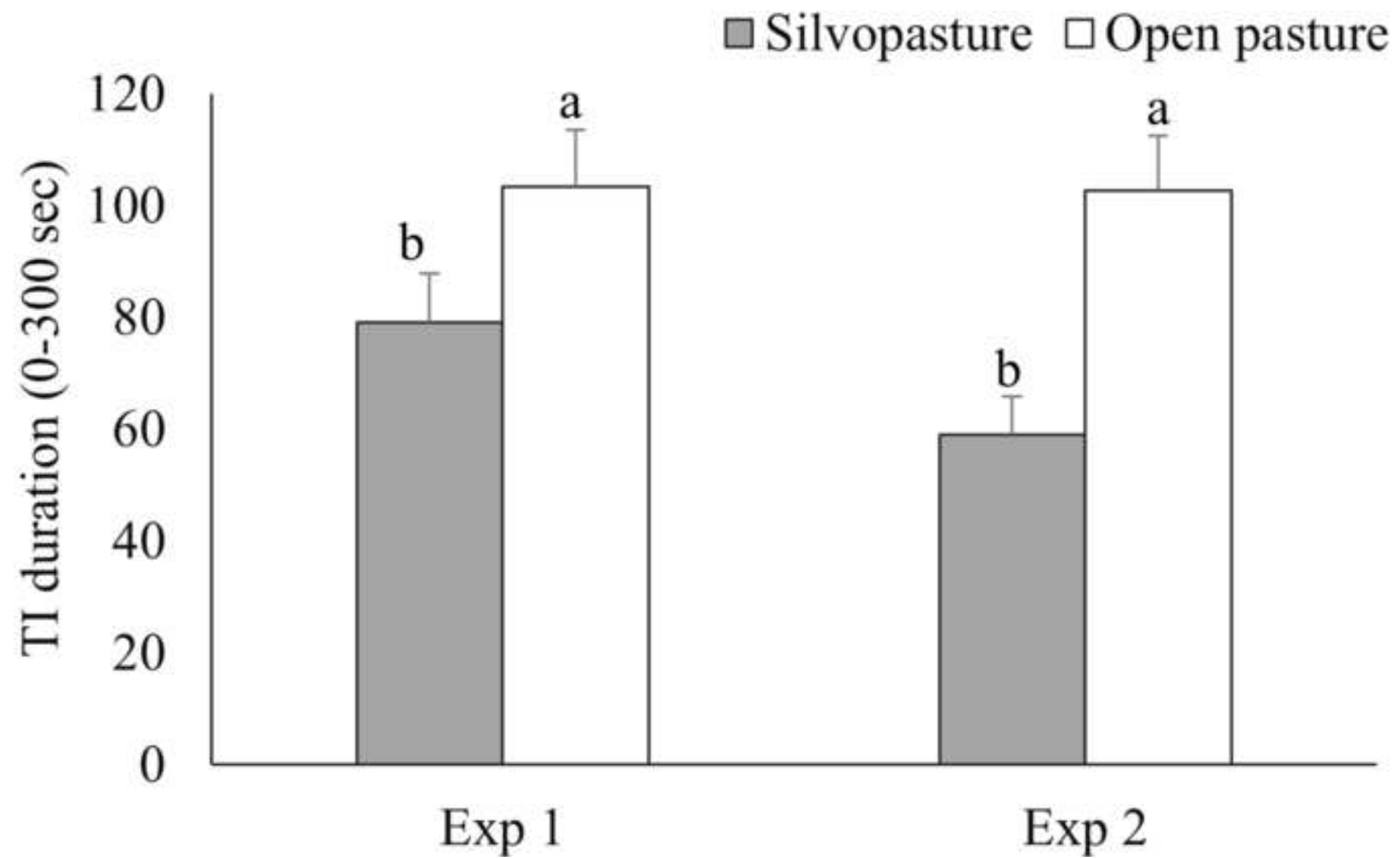
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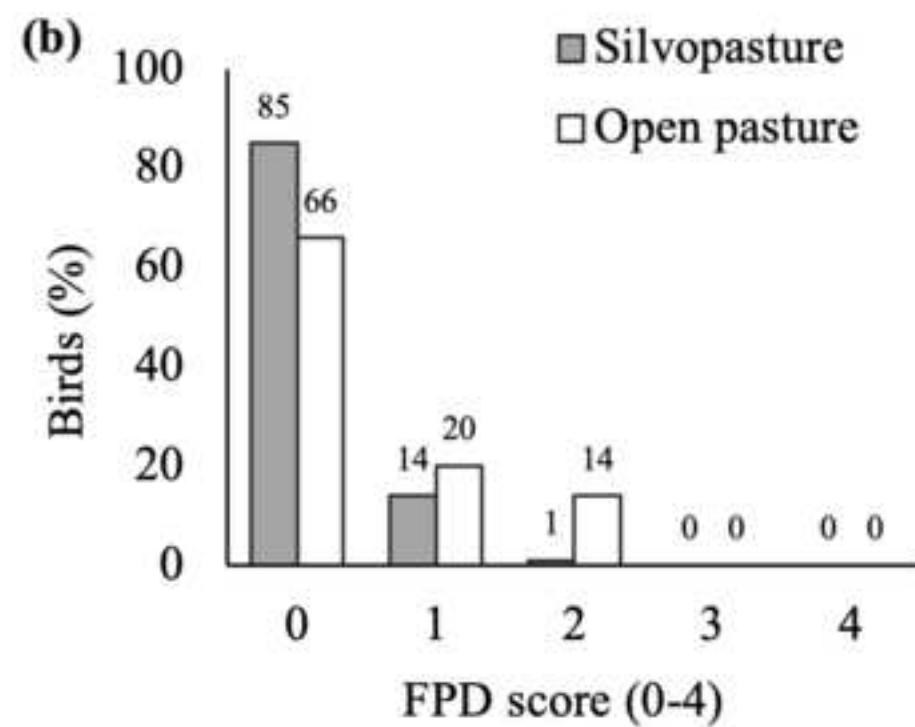
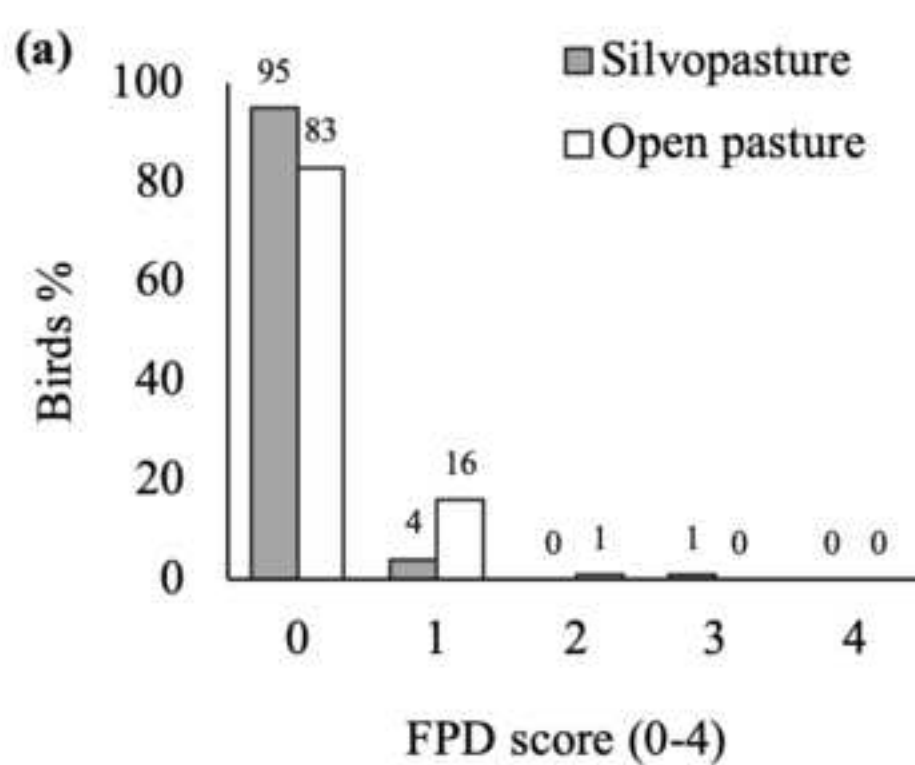
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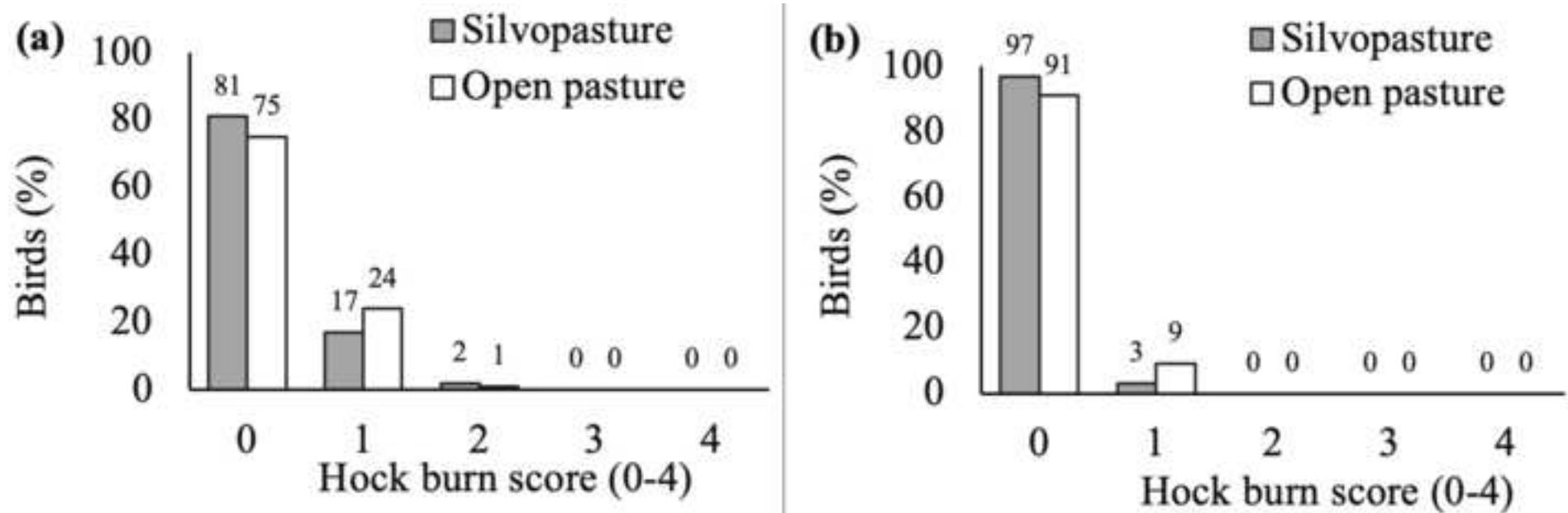
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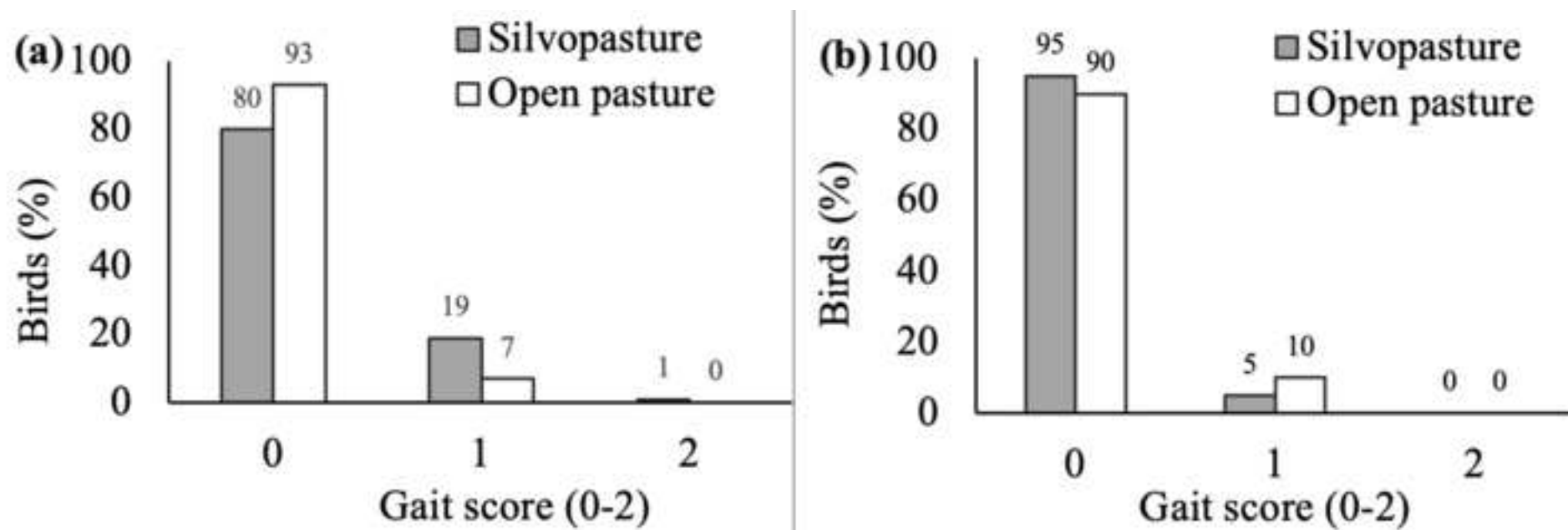
Figure 1











21 **Abstract**

22 A silvopasture system intentionally integrates trees, forages, and livestock, allowing dual
23 land use. These systems can provide high-quality habitat for broiler chickens; however, such
24 systems have not been widely adopted by the broiler industry in the United States. The objective
25 of this study was to examine the effect of silvopasture versus open pasture access on fearfulness
26 and leg health in fast-growing broiler chickens. A total of 886 mixed-sex Ross 708 chicks in
27 Experiment 1 (Exp 1) and 648 chicks in Experiment 2 (Exp 2) were housed in coops and had
28 access to 16 (Exp 1) or 12 (Exp 2) 125m² silvopasture plots (\bar{x} = 32% canopy cover) or open
29 pasture plots (no canopy cover) from day 24 of age. Fearfulness was measured using a tonic
30 immobility test (tonic immobility duration), and leg health was assessed by quantifying footpad
31 dermatitis, hock burns, gait, and performing a latency-to-lie test on days 37-39 of age. Birds in the
32 silvopasture treatment were less fearful than birds in the open pasture treatment. Overall, birds in
33 both silvopasture and open pasture systems had excellent leg health. Silvopasture birds had lower
34 footpad dermatitis scores than open pasture birds. ~~Silvopasture birds had or tended to have~~
35 ~~healthier hocks than open pasture birds.~~ Silvopasture birds ~~had tended to have~~ worse gait than open
36 pasture birds in Exp 1, but not in Exp 2. ~~Hock burn scores and~~ Latency-to-lie did not differ
37 between treatments in Exp 1 or Exp 2. Raising birds in silvopasture reduced fear and improved
38 footpad ~~and hock~~ health compared to birds raised in open pastures, which indicates that
39 silvopasture systems provide some benefits for affective state and leg health in fast-growing
40 broilers.

41
42 **Keywords:** Agroforestry, animal welfare, broiler chicken, fearfulness, leg health, outdoor
43 access

44

45 **Introduction**

46 The United States (US) has three main broiler chicken production systems which mandate
47 that birds have outdoor access. The first is certified organic production overseen by the United
48 States Department of Agriculture (USDA), which requires that birds have access to the outdoors,
49 with shade, shelter, exercise areas, fresh air, direct sunlight, and protection from predators year-
50 round [1]. The second is free-range poultry production, overseen by the USDA, where producers
51 must meet the required housing conditions to be able to sell products using the free-range label
52 [2]. These conditions include continuous free access to the outdoors for more than 51% of the
53 animals' lives [2]. The third production system is pastured poultry production, which could be
54 certified by a number of voluntary assurance schemes. Depending on those assurance schemes
55 requirements for outdoor access can vary. Some animal-welfare benefits of these systems
56 compared to indoor-only housing systems include more space and more opportunities to perform
57 natural behaviors [3–6].

58 Broiler chicken production with outdoor access requires more land than indoor-only
59 systems. Furthermore, birds may have negative experiences accessing the pasture when faced with
60 extreme temperature fluctuations, presence or threat of predators, and exposure to diseases and
61 parasites, compared to indoor-only conditions [7–9]. These conditions can increase production
62 costs and may lead to decreased gain compared with birds in indoor housing. For instance, lower
63 final body weights and higher feed conversion ratios were found for broilers with outdoor access
64 compared to indoor only fast-growing birds [10]. To compensate for the additional land costs as
65 well as potential impacts on animal performance, land productivity can be increased by utilizing a

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66 silvopasture system. A silvopasture system is an actively managed tree-forage-livestock system on
67 a single piece of land [11,12]. Short-term income can be generated through the production of
68 poultry products, and long-term income can be obtained through tree or forage production. In
69 addition, fruits and browse can have value for consumption by humans or broilers, which further
70 helps to improve short-term economic returns by reducing food or feed costs [11]. Along with
71 these economic benefits, silvopasture systems can reduce environmental stressors and serve as
72 high-quality habitat for broiler chickens compared to open pasture. Yet, silvopasture systems have
73 not been widely adopted by the broiler chicken industry.

74 In current production systems that allow outdoor access, ranges commonly consist of open
75 grassland habitats [6]. Grass pastures typically do not provide birds opportunities to seek cover
76 from aerial predators [10,13]. Chickens prefer trees and shrubs because they provide natural cover
77 to hide [14]. Access to pasture with trees or hedgerows can prevent predation loss, provide a milder
78 microclimate, and improve productivity and physiological health. Olive trees or sorghum in the
79 range resulted in no or <1% mortality due to predation for male naked-neck chickens, compared
80 to 2.4-2.8% mortality in flocks kept in open grass pastures [15]. The microclimate in a mature-
81 pine silvopasture system was milder compared to an open pasture system nearby in Florida, US
82 [16]. Similarly, mean wet-bulb globe temperatures were lower in silvopasture than open pastures
83 on the same farm in Virginia, US [17]. Access to a pasture with a mature hedgerow compared to
84 an open pasture resulted in improved weight gain without increasing feed costs in fast-growing
85 broilers [18].

86 Access to a pasture with trees rather than no or artificial shelters could potentially benefit
87 birds' emotional reactivity, such as fearfulness. Stadig and colleagues [19] found that far-ranging
88 slow-growing broilers were less fearful with access to willows than close-ranging broilers that had

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89 access to artificial shelters. The complex environment of a silvopasture may reduce fearfulness
90 similarly as indoor-only environments with enrichment objects [20,21]. For instance, broilers or
91 broiler breeders were less fearful when housed with perches [22,23], music [234], dust baths and
92 temporary objects [21] such as balls, strings, or wall drawings [245,256] compared to broilers
93 without enrichments. The potential benefit for reducing fearfulness in fast-growing broilers with
94 silvopasture access compared to open pastures has not yet been studied, even though these fast-
95 growing broilers are the common strains used in certified-organic and some free-range and
96 pastured poultry production systems in the US.

97 In conventional indoor-only broiler chicken production systems, animal welfare concerns
98 include poor leg and foot health. In a heavy-bird (~3.7 kg) indoor-only production system in the
99 US, 47.5% of birds had superficial or deep footpad lesions [267]. These lesions, referred to as
100 footpad dermatitis (FPD), are problematic from an economic [278,289] and animal welfare
101 perspective [2930]. Broilers can experience pain associated with FPD [304] and will be less
102 motivated to access feeders and drinkers. These animals gain less weight than broilers without
103 FPD and thus have lower carcass yields [2930,312,323]. Providing broilers with outdoor access
104 can reduce the prevalence of FPD. The outdoor space reduces in-house stocking density, increasing
105 birds' space, and thereby reducing point contamination with feces and moisture within the house.
106 For instance, higher litter moisture levels were found in a barn system (38% vs. 28%) compared
107 to a free-range system for turkeys between 18 and 20 weeks of age [334]. The lower litter moisture
108 concentrations indoors reduce the chances of developing contact dermatitis, including FPD, breast
109 dermatitis and hock burn lesions [2930,323,345,356]. Access to an outdoor range would therefore
110 reduce chances of FPD and hock burn lesion development in two ways, (1) by distributing fecal
111 material across the range (thus decreasing fecal material deposition in house) and (2) by reducing

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112 birds' time sitting inside, which could lessen their contact with moisture and feces. A silvopasture
113 system may encourage even greater range utilization compared to an open pasture in fast-growing
114 broilers. For example, range use in medium-growing broilers was improved with access to conifer
115 wigwams compared to birds in open pasture [367]. Similarly, range use in slow-growing broilers
116 with access to short rotation coppice was 7% greater compared to birds with artificial shelters in
117 the range [378]. In commercial operations (indoor-only, heavy-bird production) in the US, 34.5%
118 of birds had abnormal gait or were totally lame [267]. Over 27% of indoor-only housed broilers
119 had poor walking ability in the United Kingdom, with 3% of birds having limited ability to walk
120 at 40 days of age [389]. Outdoor access could improve gait and reduce lameness. Slow-growing
121 broilers that used their outdoor range had improved gait compared to birds that were indoors [3940]
122 or used the range less [19]. Yet, the potential benefit of silvopasture access for improving leg health
123 in fast-growing broilers has not been studied.

124 Several studies have shown animal welfare benefits of outdoor access for fast-growing
125 broilers [18,404], slow-growing broilers [19,3940,412,423], and turkeys [334]. However, it is
126 unclear whether the welfare benefits of pasture access, and especially silvopasture access, can be
127 obtained for fast-growing broiler chickens, which are the most common type of broiler chicken
128 produced in the US. Therefore, our study objective was to evaluate the effect of silvopasture versus
129 open pasture access on fearfulness and leg health in fast-growing broiler chickens. We
130 hypothesized that birds in silvopasture systems would be less fearful and have improved leg health
131 (FPD, hock, gait, and latency to lie) than birds in open pasture systems.

132 **Materials and Methods**

133 **Animals and housing**

134 Two experiments were conducted from April to May (Experiment 1; Exp 1) and June to
135 August 2021 (Experiment 2; Exp 2). All procedures were approved by the Virginia Tech
136 Institutional Animal Care and Use Committee (IACUC protocol 20-044).

137 In total, 886 one-day-old Ross 708 mixed-sex chicks in Exp 1 and 648 chicks in Exp 2
138 were obtained from a commercial hatchery (Harrisonburg, VA, USA). Birds were Marek's
139 vaccinated at the hatchery. Upon arrival, chicks were arbitrarily selected and housed in 12 identical
140 pens (5.7 m²) with 73 or 74 birds per pen in Exp 1 and 53 or 54 birds per pen in Exp 2. Pens
141 contained pine wood shavings (~5 cm depth), a heat lamp (day 1-8), a cardboard feed flat with
142 feed (day 1-8), one bell drinker (Plasson® Broiler Drinker complete, Or-Akiva, Israel), and one
143 pet champion poultry drinker (Stout Stuff LLC, China), and one feeder (Superbowl poultry feeder,
144 LaGrange, NC, USA). The chicks were fed commercial starter (day 0-15), grower (day 15-25),
145 and finisher (day 25-42 or 43) diets meeting requirements [434]. Ambient temperatures were 35°C
146 on day 1 and gradually reduced to 23°C on day 22 (Exp 1) or day 23 (Exp 2). Lighting was
147 provided continuously for the first week and reduced to 12h light and 12h dark until day 22 (Exp
148 1) or day 23 (Exp 2).

149 In Exp 1, birds from each pen were equally and randomly allocated over 16 pasture-based
150 treatments resulting in 53 birds per plot. In Exp 2, complete pens (53-54 birds) were randomly
151 allocated to 12 pasture-based plots with chicken coops. On day 22 (Exp 1) or day 23 (Exp 2), birds
152 were transported for 1.5h to the pasture-based experimental site located at the Shenandoah Valley
153 Agricultural Research and Extension Center (AREC) in Raphine, VA, US. After transportation,
154 birds were kept inside the coops for two days (Exp 1) or one day (Exp 2) to get acclimated to their

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155 new housing conditions. From day 24, coop doors in each plot were opened at approximately 8:00
 156 AM and closed at approximately 5:00 PM.

157 All pasture-based plots (125m²) contained a chicken coop (6.55m²) constructed from wood,
 158 chicken wire, and tarp [445]. Each coop contained a wooden platform perch (0.05m × 0.10m ×
 159 2.40m), feeder, and bell drinker. Coops were moved laterally across the plot each week. Plots were
 160 fenced with 1-m-high and 50-m-long FlexNet electric fences (PoultryNet®, Washington, IA,
 161 USA), connected to a 30-volt electric cattle fence. Mean pen and coop stocking densities on day
 162 1, day 22 or 23, and day 42 or 43 for Exp 1 or Exp 2 are given in Table 1.

163
 164 **Table 1. Mean (± SEM) of pen and coop stocking density on day 1, day 22-23, and day 42-43**
 165 **of age in Experiments 1 (Exp 1) and 2 (Exp 2).**

Stocking Density	Pen		Coop			
	Day 1		Day 22		Day 42	
	g/m ²	Birds/m ²	kg/m ²	Birds/m ²	kg/m ²	Birds/m ²
Exp 1	518.1±0.8	13.0±0.0	7.5±0.1	8.0±0.0	20.8±0.4	7.2±0.1
	Day 1		Day 23		Day 43	
Exp 2	439.3±13.3	9.4±0.0	7.4±0.1	8.2±0.0	21.8±0.2	8.1±0.1

166
 167 **Treatments**

168 The silvopasture plots (four replicates in two locations in Exp 1, and three replicates in two
 169 locations in Exp 2; Fig 1) contained a mixed hardwood stand of black walnut (*Juglans nigrea* L.),
 170 locust (*Robinia pseudoacacia* L.) and hickory (*Carya* spp. Nutt.) trees and 30 newly planted

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171 saplings per plot (American hazelnut (*Corylus americana*), black walnut (*Juglans nigra*),
172 persimmon (*Diospyros virginiana* L.), southern red oak (*Quercus falcata* Michx.), and southern
173 pine (*Pinus* spp.) of approximately 30-cm height and 1-cm diameter. Saplings were planted in six
174 rows with inter- and intra-row spacing of 1.5m. Canopy cover in all silvopasture and open pasture
175 plots was calculated from photos using ImageJ software (1.5.3k, National Institutes of Health,
176 Bethesda, MD, USA). The images were taken straight upwards from ground level in the center of
177 the plot. Photos were converted to 8-bit, binarized, and then the number of black (canopy) and
178 white (sky) pixels were calculated as a percentage of total pixels. The canopy cover for silvopasture
179 plots was (mean±standard deviation) 31.7±16.7% in Exp 1, and 33.3±10.9% in Exp 2. Open
180 pasture plots had no canopy cover.

181 The open pasture plots (four replicates in two locations in Exp 1 and three replicates in two
182 locations in Exp 2; Fig 1) contained ground vegetation. Common herbaceous forage species in the
183 open pasture and silvopasture plots were tall fescue (*Schedonorus arundinaceus* (Schreb.)
184 Dumont., syn. *Lolium arundinaceum* (Schreb.) Darbysh., formerly *Festuca arundinacea* Schreb.),
185 orchard grass (*Dactylis glomerata* L.), horse nettle (*Solanum carolinense* L.), and common
186 milkweed (*Asclepias syriaca* L.). Forage species specific to silvopasture plots includes greenbrier
187 (*Smilax* spp. L.), honeysuckle (*Lonicera japonica* Thunb.), Virginia creepers (*Parthenocissus*
188 *quinquefolia* L. Planch.), nimble will (*Muhlenbergia schreberi* J.F. Gmel.), poison ivy
189 (*Toxicodendron radicans* (L.) Kuntze ssp. *radicans*), wild basil (*Clinopodium vulgare* L.),
190 common blue violet (*Viola sororia* Willd), and wood sorrel (*Oxalis* spp.). Forage species specific
191 to open pasture plots were common chickweed (*Stellaria media* (L.) Vill.), common dandelion
192 (*Taraxacum officinale* F.H. Wigg.), and Queen Anne's lace (*Daucus carota* L.).

193

194 **Fig 1. ~~Google Maps image~~ and illustration of experimental plots (125m² each).** Open
195 grassland pasture plots (~~red~~orange) and silvopasture plots (black/white) contained a chicken coop
196 (black). Experiment 1 included 4 replicates per treatment in two locations, and Experiment 2
197 included 3 replicates per treatment in two locations. The omitted replicate plots for Experiment 2
198 are marked with a pattern fill. [Map sourced from USGS National Map Viewer.](#)

200 **Measurements**

201 A total of 79 or 80 birds per treatment (159 birds total) in Exp 1 and 78 birds per treatment
202 (156 birds total) in Exp 2 received a numbered leg band on day 37 or 38 of age for individual
203 identification. In Exp 1, 10 birds per plot were sampled; in Exp 2, 13 birds per plot were sampled.
204 All measurements were performed on the same birds. [As sampling took place on pasture, observers](#)
205 [could not be blinded to experimental treatments.](#)

207 **Tonic immobility**

208 Birds were tested for tonic immobility (TI) duration and TI induction on day 38 in Exp 1
209 and day 37 of age in Exp 2. Two observers performed the TI test in both experiments. The inter-
210 observer agreement was tested for 10 birds and was excellent (Cronbach's α of 0.92). The TI test
211 was conducted as described by [21]. However, birds were tested outdoors in the pasture rather than
212 indoors. During the TI test, the assessed birds did not have visual contact with other birds or the
213 observer. A bird was placed on its back in a V-shaped cradle, and then TI was induced by
214 restraining the bird with one hand on its sternum for 15 seconds while covering their head with the
215 other hand. At the end of the induction period, both hands were gently removed. If the bird tried
216 to right itself within 10 seconds, the induction attempt was considered failed, and the handler

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217 repeated the restraint procedure (no more than three induction attempts). In Exp 1, one bird for
218 which TI could not be induced was replaced with another bird. In Exp 2, birds that could not be
219 induced were included in the sample and received a TI duration of 0 seconds. After successful
220 induction of TI, the TI duration was recorded for a maximum of 5 minutes.

221

222 **Footpad dermatitis and hock burn lesions**

223 Eighty birds per treatment (160 birds total) in Exp 1 and 77 or 78 birds per treatment (155
224 birds total) in Exp 2 were assessed for FPD and hock burn lesions on day 39 in Exp 1 and day 38
225 in Exp 2. FPD and hock burn lesions were scored on a 0-4 categorical scale, with increasing scores
226 indicating worse lesions [456]. A single trained observer scored FPD and hock burn lesions in Exp
227 1 and Exp 2 and recorded the most severe score of a bird's two feet or hocks.

228

229 **Gait**

230 Birds were evaluated individually for their walking ability and assigned a categorical gait
231 score between 0-2, with higher scores representing worse gait [467]. A single trained observer
232 performed the scoring by voluntarily allowing the birds to walk for at least 1.5m. If the birds did
233 not walk, the observer stimulated the bird by gently touching their tail or vent with a rod. If the
234 bird did not walk after gentle stimulation, the bird received the highest gait score (score 2).

235

236 **Latency to Lie**

237 Fifty-five or 56 birds per treatment (111 birds total) in Exp 1 and 77 birds per treatment
238 (154 birds total) in Exp 2 were individually assessed in a latency to lie (LTL) test on days 39 or
239 40 in Exp 1 and days 37 or 38 in Exp 2. The test was performed as described by [478]. The LTL

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240 was recorded as an indicator of leg strength, with shorter latencies representative of poorer leg
241 strength [478]. Individual birds were placed in an opaque plastic tub (0.93m L × 0.54m W × 0.46m
242 H; Sterilite Corporation, Townsend, MA, USA) containing 2-3cm lukewarm (29 - 36°C) water for
243 a maximum of 10 minutes. In Exp 2, the tub was covered with a barrier made from bird netting
244 and PVC pipe to avoid escape behaviors observed in Exp 1. Four birds were tested simultaneously
245 in separate tubs. Birds that remained standing at the end of the test received an LTL of 10 minutes.
246 A single observer performed the tests.

247

248 **Weather and soil conditions**

249 Ambient temperature (°C), photosynthetically active radiation ($\mu\text{Em}^{-2}\text{s}^{-1}$), soil moisture (%
250 volumetric moisture content), relative humidity (%), and dew point (°C) were measured using one
251 Spectrum WatchDog 1000 Series MicroStation (Spectrum Technologies, Inc., Aurora, IL, USA)
252 per treatment from day 27 to day 43 in Exp 1.

253

254 **Statistical analysis**

255 Data of both experiments were analyzed separately in JMP Pro 16 (SAS Institute Inc., Cary,
256 NC, USA). Data residuals of continuous response variables were assessed for the normality of
257 their distribution by visual inspection of normal quantile plots. Data residuals of TI duration ~~and~~
258 ~~TI induction attempts, FPD, hock, gait,~~ and LTL were not normally distributed. TI duration was
259 log-transformed and analyzed using a mixed model with treatment as fixed factor and plot as
260 random factor. TI attempts (counts) were analyzed in SAS Studio 3.8 (SAS Institute Inc., Cary,
261 NC, USA) with a linear mixed model with a Poisson distribution, using treatment as fixed factor
262 and plot as random factor. Transformation of LTL did not result in normality of residuals; thus,

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263 ~~plot means were calculated to obtain a single measure per experimental unit and avoid pseudo-~~
264 ~~replication. These were analyzed in a mixed model with treatment as fixed factor and plot as~~
265 ~~random variable. Ordinal response variables (FPD, hock, and gait scores) were analyzed in SAS~~
266 ~~Studio using generalized linear mixed models with a multinomial (ordered) distribution, with~~
267 ~~treatment as fixed factor and plot as random factor. ,thus data were analyzed using non-parametric~~
268 ~~tests. Outliers were not removed from the dataset. Wilcoxon tests were used to analyze continuous~~
269 ~~variables (TI duration and LTL) with pasture treatment as a fixed factor. Ordinal logistic models~~
270 ~~were used to analyze ordinal variables (TI induction attempts, FPD, hock, and gait scores) with~~
271 ~~pasture treatment as a fixed factor.~~ The threshold for significance was set at $p \leq 0.05$ and for a trend
272 at $p \leq 0.10$. Raw means and standard errors are reported unless otherwise noted.

274 Results

275 Tonic immobility

276 Silvopasture birds ~~tended to showed~~ shorter TI durations than open pasture birds in Exp 1
277 ~~($F_{1,14}=3.89$, $p=0.069$) and were shorter and in Exp 2 ($F_{1,10}=24.04$, $p<0.001$ ($\chi^2=4.668$; $p=0.031$ in~~
278 ~~Exp 1, and $\chi^2=15.993$; $p<0.001$ in Exp 2; Fig 2). No difference was found ($\chi^2=0.496$; $p=0.481$) in~~
279 the number of attempts to induce TI among birds in the silvopasture and open pasture treatments
280 in Exp 1 ($F_{1,143}=0.19$; $p=0.664$) or Exp 2 ($F_{1,143}=0.73$; $p=0.395$). ~~In Exp 2, silvopasture birds~~
281 ~~tended to require fewer attempts (1.2 ± 0.1 vs. 1.5 ± 0.1 attempts) to induce TI than open pasture~~
282 ~~birds ($\chi^2=3.356$; $p=0.067$).~~

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284 **Fig 2. Means for tonic immobility (TI) duration (\pm SEM) for broilers raised in silvopasture**
285 **or open pasture plots on day 38 in Experiment 1 (n=159) and on day 37 of age in Experiment**
286 **2 (n=156).** ^{ABab} Raw means within ~~experiment-Experiment 1(Exp 1 or Exp 2)~~ with uncommon
287 superscripts differ at $p < 0.05$. ^{ab} Raw means within Experiment 2 with uncommon superscripts
288 differ at $p < 0.05$.
289

290 Leg health

291 Silvopasture birds ~~tended to have~~ showed lower (improved) FPD scores than open pasture
292 birds in Exp 1 ($F_{1,141}=3.30$; $p=0.071$) and had lower scores in Exp 2 ($\chi^2=13.463$; $p=0.001$)
293 ($F_{1,143}=5.61$; $p=0.019$ in Exp 1 and $\chi^2=8.515$; $p=0.004$ in Exp 2; Fig 3). Mean FPD scores were
294 0.08 \pm 0.04 (Exp 1) and 0.17 \pm 0.05 (Exp 2) for silvopasture birds, and 0.19 \pm 0.05 (Exp 1) and
295 0.48 \pm 0.08 (Exp 2) for open pasture birds. ~~In Exp 1, silvopasture birds showed lower (improved)~~
296 ~~hock burn scores than open pasture birds ($\chi^2=3.853$; $p=0.050$)~~ Hock burn scores did not differ
297 between treatments in Exp 1 ($F_{1,142}=2.14$; $p=0.146$) or Exp 2 ($F_{1,143}=2.66$; $p=0.105$; Fig 4). ~~In~~
298 ~~Exp 2, silvopasture birds tended to have lower hock burn scores than open pasture birds ($\chi^2=3.182$;~~
299 ~~$p=0.074$; Fig 4).~~ Mean hock burn scores were 0.15 \pm 0.05 (Exp 1) and 0.03 \pm 0.02 (Exp 2) for birds
300 in the silvopasture treatment, and 0.26 \pm 0.05 (Exp 1) and 0.09 \pm 0.03 (Exp 2) for birds in the open
301 pasture treatment. In Exp 1 but not in Exp 2, silvopasture birds ~~tended to have~~ had worse gait scores
302 than open pasture birds ($F_{1,143}=2.96$; $p=0.088$) ($\chi^2=5.517$; $p=0.019$ in Exp 1 and $F_{1,143}=1.44$;
303 $p=0.231$ $\chi^2=1.820$; $p=0.117$ in Exp 2; Fig 5). The majority of sampled birds showed excellent gait,
304 with $\geq 80\%$ of birds having a gait score 0 (Fig 5). Mean gait scores were 0.21 \pm 0.05 (Exp 1) and
305 0.05 \pm 0.03 (Exp 2) for silvopasture birds, and 0.08 \pm 0.03 (Exp 1) and 0.10 \pm 0.04 (Exp 2) for open
306 pasture birds. No difference was found in LTL between silvopasture birds and open pasture birds

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307 in Exp 1 ($F_{1,14}=1.04$, $p=0.324$) and Exp 2 ($F_{1,10}=1.22$, $p=0.295$) ($\chi^2=1.493$; $p=0.222$ in Exp 1 and
308 $\chi^2=0.196$; $p=0.657$ in Exp 2). The mean LTL for silvopasture and open pasture birds was 452 ± 28
309 sec (Exp 1) and 523 ± 19 sec (Exp 2) for birds in silvopasture, and ~~vs.~~ 403 ± 28 sec ~~in~~ (Exp 1); and
310 ~~523±19 sec vs.~~ 549 ± 13 sec ~~(in~~ Exp 2) for birds in open pasture.

311
312 **Fig 3. Percentage of birds with footpad dermatitis (FPD) scores 0-4 in the silvopasture and**
313 **open pasture treatments.** (a) On day 39 of age in Experiment 1 ($n=160$). (b) On day 38 of
314 age in Experiment 2 ($n=155$).

315
316 **Fig 4. Percentage of birds with hock burn scores 0-4 in the silvopasture and open pasture**
317 **treatments.** (a) On day 39 of age in Experiment 1 ($n=160$). (b) On day 38 of age in Experiment
318 2 ($n=155$).

319
320 **Fig 5. Percentage of birds with gait score 0-2 in the silvopasture and open pasture treatments.**
321 (a) On day 39 of age in Experiment 1 ($n=160$). (b) On day 38 of age in Experiment 2 ($n=155$).

323 Weather and soil conditions

324 Ambient temperature, photosynthetically active radiation, soil moisture, relative humidity,
325 and dew point in the silvopasture and open pasture treatments in Exp 1 are presented in Table 2.
326 Although not statistically analyzed, all but one (soil moisture) values were lower in the silvopasture
327 treatment compared to the open pasture treatment.

329 **Table 2. Raw means (\pm SEM) of weather and soil conditions in the silvopasture and open**
 330 **pasture treatments from day 27 to day 43 in Experiment 1 (n=2).**

Weather and soil conditions	Silvopasture	Open pasture
Ambient temperature ($^{\circ}$ C)	14.1 \pm 0.1	15.0 \pm 0.2
Photosynthetically active radiation (μ Em ⁻² s ⁻¹)	171.4 \pm 4.6	272.4 \pm 6.7
Soil moisture (% volumetric moisture content)	99.8 \pm 0.1	16.8 \pm 0.2
Relative humidity (%)	64.8 \pm 0.3	70.9 \pm 0.2
Dew point ($^{\circ}$ C)	6.5 \pm 0.1	8.0 \pm 0.1

331

332 Discussion

333 This study investigated the effect of silvopasture versus open pasture access on fearfulness
 334 using a TI test and on leg health using FPD scores, hock burn scores, gait scores, and LTL in fast-
 335 growing broiler chickens. We found that pasture type impacted fearfulness, FPD, ~~hock burn~~
 336 ~~lesions~~, and tended to impact gait scores, with silvopasture access showing improvements for all
 337 measures besides gait, and no difference in hock burns or LTL.

338

339 Tonic immobility

340 The shorter TI duration in birds in the silvopasture treatment in Exp 1 and Exp 2 indicates
 341 that those birds were less fearful than birds in the open pasture treatment [489]. TI durations in the
 342 current study were shorter than durations reported in previous research with conventional broilers
 343 raised in different housing conditions; indoor-only [10,423,4950,504], indoor housing with
 344 outdoor access [10,4950-512], and indoor housing with environmental enrichment [10,245,523-

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345 545]. TI durations (103-104 sec) in our open pasture treatments were comparable to the duration
346 of a slow-growing broiler strain housed with outdoor access (108 sec; [423]). Far-ranging slow-
347 growing broilers were less fearful with access to willows than close-ranging broilers with access
348 to artificial shelters [19], suggesting that trees in the range were more meaningful to reduce
349 fearfulness than artificial shelters in their study. This is in line with findings in our study, as we
350 also found birds with access to trees to be less fearful than birds without trees in the range. Lack
351 of overhead cover in open pasture plots could have increased the birds' chance of encountering
352 predators, increasing broilers' fear and anxiety [556] compared to birds in the silvopasture plots.
353 In the current study no mortality from areal predation occurred, yet one account of predation by a
354 ground predator was observed in an open pasture plot (Exp 1), thought to be caused by a raccoon
355 (*Procyon lotor*), when birds were 42 days of age, which was 4 days after fear was assessed.

356 In addition to being obscured from predators, we argue that a more complex outdoor range
357 with trees could serve a similar function as enrichments provided when housed indoors, either
358 biologically relevant (perches) or biologically less relevant (such as music or balls), as both
359 enrichments and access to a complex range promote the expression of natural behaviors
360 [3,5,567,578]. Broilers raised with environmental complexity in indoor-only housing conditions
361 were less fearful compared to broilers housed in simpler conditions. For instance, Arbor Acres
362 birds with music [234] and Ross 708 birds with dust baths, perches, and temporary objects [21]
363 were less fearful than without enrichments. Our results indicate that allowing fast-growing broiler
364 chickens access to a silvopasture range reduces fearfulness compared to access to an open pasture
365 range.

366

367 **Footpad dermatitis and hock burn lesions**

368 Broilers with access to silvopasture had better FPD scores than broilers with access to open
369 pasture. Five percent of birds in Exp 1 and 15% of birds in Exp 2 had mild or severe FPD scores
370 in the silvopasture treatment, which is lower or comparable to reported prevalences of 12% and
371 22% in slow-growing broilers with access to olive trees or sorghum [412], and lower than
372 prevalences of 68% and 72% in slow-growing broilers with access to willows or artificial shelters
373 [19]. Litter (soil) moisture content is identified as a key risk factor for the development of contact
374 dermatitis [323,356,589]. The lower FPD prevalence and severity in silvopasture plots, even with
375 higher soil moisture content (Table 2), may be due to increased range use in silvopasture plots
376 compared to open pasture plots, which could have reduced the in-coop stocking density, reduced
377 fecal material within the coop, and reduced birds' time sitting inside. Despite the soil moisture
378 content being higher in the silvopasture treatment compared to the open pasture treatment, our
379 findings show that FPD prevalence and severity in fast-growing broilers are minor in either pasture
380 system, but especially minimal in the silvopasture system. Although not statistically assessed, FPD
381 scores were lower in Exp 1 compared to Exp 2. Lower maximum in-coop stocking density in Exp
382 1 versus 2 (Table 1) could have contributed to that difference. Seasonal variation in FPD scores in
383 fast-growing indoor-only broilers was reported, with the best outcomes during the warmest months
384 [59], yet those results do not align with our findings, as FPD was slightly worse in summer
385 compared to spring.

386 Hock health was excellent in birds with access to either open pasture or silvopasture and
387 did not differ between treatments. Similar to FPD scores, severity and prevalence of hock burns
388 were low in both treatments, ~~with healthier hocks when broilers had access to silvopasture plots~~
389 ~~compared to open pasture plots~~. Although no previous work has investigated hock burn lesions in

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390 fast-growing broilers with pasture access, our findings generally do align with earlier research on
391 slow-growing broilers with outdoor access [40+,423]. Hock burn scores for silvopasture and open
392 pasture birds in the current study were better when scored on a 5-point scale (mean scores between
393 0.03-0.26; ~~data not shown~~) than reported for slow-growing broilers from seven organic farms with
394 outdoor access scored using a similar 4-point scale (mean score of 0.30; [423]). Birds in the
395 silvopasture treatment had comparable or higher hock burn scores as slow-growing broilers from
396 25 farms with outdoor access (score 0=96% and score 1=4%; [40+]), while birds in the open
397 pasture treatment showed more severe hock burn lesions compared to those reported by [40+].
398 These results met our expectations as FPD and hock burn lesions can show a positive association
399 and share some of the same etiology [60].

400

401 Gait

402 Most birds in the current study had excellent gait, with the majority showing no gait
403 imperfection in either treatment group. Outdoor access has been associated with better gait in fast-
404 growing [512,61] and slow-growing broilers [19,51] compared to indoor-only housing. Although
405 [10] found no difference in gait scores in fast-growing broilers raised indoors compared to broilers
406 with outdoor access. Even though access to pasture resulted in good gait across treatments, gait
407 ~~was tended to be worse~~ed in the silvopasture treatment compared to the open pasture treatment
408 in Exp 1 only. This is contrary to our hypothesis and does not align with the outcomes for other
409 leg health indicators that were assessed, where silvopasture resulted in an improvement or no
410 difference. The mechanism behind the difference in gait scores in Exp 1 is unclear. The lack of a
411 difference in Exp 2 suggest that it may not be consistently due to the pasture treatment, but may
412 have been a result of other factors.

413 **Latency to Lie**

414 The LTL test measures leg strength in broilers by recording the time it takes for birds to lie
415 down in lukewarm water [62], relying on chickens' aversion to sit down in water, therefore, shorter
416 latencies reflect poorer leg strength [478]. LTL did not differ between treatments in either
417 experiment. In the current study, birds showed comparable LTL, thus suggesting comparable leg
418 strength to slow-growing strains (mean LTL between 403-548 sec vs. 547 sec reported by [423]).
419 Generally, leg health is worse in fast-growing broilers compared to slow-growing strains [63]. This
420 suggests that both treatments in the current study resulted in good leg strength in fast-growing
421 broilers, which is a major achievement in terms of animal welfare, as leg issues are a well-
422 recognized welfare concern for fast-growing broilers [389,64].

423

424 **Conclusion**

425 This study evaluated the effect of silvopasture versus open pasture systems on fearfulness
426 and leg health in fast-growing broiler chickens. To our knowledge, this is the first study to assess
427 the impact of a pasture system (silvopasture or open pasture) on aspects of animal welfare and
428 health of fast-growing broiler chickens. We found that providing fast-growing broilers with access
429 to a silvopasture system from 3 weeks of age is particularly beneficial for reducing fearfulness and
430 improving leg health, specifically footpad ~~and hock~~ condition, compared to providing access to
431 open pasture. Even though gait was worse in silvopasture-raised broilers in one of two
432 experiments, scores were low (thus gait was not impaired) in either treatment. Leg strength was
433 good and comparable in both treatments.

434

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439

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