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Vliv systému ustájení na užitkovost, kvalitu masa, zdraví a welfare králíků

doktorská disertační práce

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Obsah

1 Literární přehled

1.1 Systémy ustájení

Podle autorů EFSA (2020) lze systémy ustájení rozdělit na konvenční klece, obohacené klece a vyvýšené kotce (typicky používané na konvenčních farmách), podlahové boxy, venkovní a ekologické systémy.

Konvenční klece

Tento typ klecí se používá pro vykrmované králíky, mladé a nebřezí samice. Speciálními typy konvenčních klecí jsou bicelulární klece pro chov králíků a dvouúčelové klece pro reprodukci samic s mláďaty nebo pro rostoucí králíky (EFSA, 2020). Tradičně se bicelulární klece používají jako konvenční systém ustájení pro výkrm králíků. EFSA (2020) udává velikost bicelulární klece 25,4 x 44 x 28 cm (šířka x délka x výška) s celkovou dostupnou plochou 1 200 cm^2. Xiccato a kol. (2013) doporučují 28 x 40 x 28 cm. Trocino a kol. (2013) doporučují 28 x 40 x 30 cm. Tento systém ustájení byl po velmi dlouhou dobu nejběžnějším typem ustájení na farmách králíků (Matics a kol., 2019).

Jednou z nejdůležitějších částí klece je podlaha. Nejčastějším typem podlahy je drátěná síť, v mnoha případech spárovaná s plastovou podložkou na nohy (25 x 36 cm). Běžně se také používají plastové rošty. V ustájovacím prostoru musí být přítomno krmítko a niplová napáječka. Konvenční klece slouží k ustájení pouze 2 vykrmovaných králíků (EFSA, 2020) s hustotou osazení 18 zvířat/m^2 (Xiccato a kol., 2013).

Obohacené klece

EFSA (2020) uvádí dva typy těchto klecí, obohacené klece s drátěnou podlahou (38 - 46 x 95 - 102 x 60 - 65 cm, celková dostupná plocha 4 370 - 4 700 cm^2) a obohacené klece s poličkou z plastového roštu (46 - 52,5 x 102 x 65 - 80 cm, celková dostupná plocha 5 600 - 6 400 cm^2). Tyto klece poskytují více volného prostoru pro králíky a musí být obohaceny o vyvýšené plošiny, okus nebo jiné předměty.

Vyvýšené boxy (combi-parky)

Vyvýšené boxy pro samice s mláďaty nebo pro vykrmované králíky (180 - 200 cm x 80 - 102 cm s otevřenou horní částí, celková dostupná plocha 18 000 - 25 400 cm^2) jsou sestaveny z jednotlivých modulů, které lze spojovat dohromady. K dispozici je jeden velký modul pro samici a její mláďata s odnímatelným plastovým hnízdem. Tento prostor je od prostoru klece

oddělen odnímatelnou stěnou. Po odstavení lze stěnu mezi moduly oddělit a dát vzniknout jednomu novému velkému prostoru, který se obvykle skládá ze čtyř modulů (EFSA, 2020). Rommers a kol. (2014) prezentují combi-parky jako 38 cm široké a 100 cm hluboké s otevřenou horní částí systému ustájení. Poličku o šířce 38 cm autoři umístili 25 cm nad plastovou podlahou, kotiště byla 25 cm dlouhá a 38 cm široká. Dal Bosco a kol. (2019) uvedli podobný systém a nazvali ho „kombinovaná koloniální klec“. Systém se skládal ze dvou-úrovňové porodní části s hnízdy a odnímatelných stěn. Rozměry této klece jsou 130 x 158 x 60 cm s vnějšími uzavíratelnými hnízdními budkami (39,52 x 20 x 35 cm).

Podlahové boxy

Tento typ ustájení použili Ruchti a kol. (2018) ve Švýcarsku pro samice s jejich mláďaty a následně pak pro vykrmované králíky. Průměrné rozměry kotce byly 3,5 x 2,0 m s otevřenou horní částí boxu a minimálním prostorem pro samici 1,6 m^2. V boxech byly instalovány vyvýšené plastové plošiny a hnízda (0,29 x 0,36 m), k nimž byl přístup samicím umožněn pouze po vyskočení.

Venkovní systémy

EFSA (2020) představuje tyto systémy jako tzv. „niche“ systémy s různými stacionárními (klece, boudy, výběhy) nebo mobilními systémy ustájení (obvykle klece), které mohou umožnit přístup do venkovních prostor a pastvin. Nejsou však k dispozici žádné standardy pro rozměry tohoto ustájení. Například Paci a kol. (2013) popisují svůj venkovní systém jako kolonii klecí s drátěnými podlahami o různých rozměrech těchto klecí (50 x 50 x 76 cm, 100 x 80 x 76 cm, 100 x 160 x 76 cm), které jsou situovány ve venkovní části postavené tak, aby byla poskytována zvířatům ochrana před predátory a slunečním zářením.

Ekologické systémy

Dle EFSA (2020) existuje nařízení EU 2018/848, které obsahuje základní požadavky jako je možnost pastvy, kdykoli to podmínky dovolí, skupinové ustájení, přístup do krytého přístřešku včetně tmavého úkrytu, vyvýšená plošina a hnízdní materiál pro všechny březí samice před porodem, žádná antibiotika a žádné hormony. Tato implementace požadavků vstoupila v platnost od roku 2021. Podmínky ekologického chovu králíků podléhají předpisům každé země. Pla (2008) chovala skupinu králíků na místní ekologické farmě podle doplňkových norem R (CEE) N° (1804/1999) CRAE (Národní regulační výbor pro ekologické zemědělství)

ve Španělsku. Používaly se skupinové kotce (2 m^2) a slaměná podestýlka, králíci byli krmeni ad libitum.

1.2 Vliv systému ustájení na produkční parametry králíků

Růst, průměrný denní přírůstek, konverze nebo příjem krmiva jsou některé z důležitých parametrů výkrmu, které mají vážný dopad na produkci králičího masa. Několik studií zkoumalo vliv systémů ustájení na produkční parametry s průkazně vyšším průměrným denním přírůstkem a konečnou živou hmotností u králíků chovaných v klecích než u králíků umístěných ve výbězích při malých skupinách (Lambertini a kol., 2001; Princz a kol., 2009) nebo u větších skupin (Dal Bosco a kol., 2002; Combes a kol., 2010).

Velikost skupiny

Při porovnání různé velikosti skupiny králíků (6, 12, 18, 30, 42, 54) nebyl zaznamenán žádný vliv na růst a příjem krmiva (Rommers a Meijerhof, 1998). Na druhou stranu, Xiccato a kol. (1999) zjistili, že denní příjem krmiva významně poklesl ve skupinách králíků v boxech ve srovnání s králíky, kteří byli chováni v klecích. Bylo také zjištěno, že pohybová aktivita se zvyšuje se zvyšující se plochou prostoru v ustájovacím systému, a negativně ovlivňuje příjem krmiva (Rommers a Meijerhof, 1998). Také králíci chovaní ve velkých skupinách vykazovali nižší příjem krmiva v důsledku vyšší úrovně stresu a agresivity (Maertens a Van Herck, 2000). Naopak Matics a kol. (2018) nezjistili žádný vliv systému ustájení na příjem krmiva, když porovnávali boxy a klece. Související vyšší pohybová aktivita ve větších skupinách pak dle více studií (Lambertini a kol. 2001; Dal Bosco a kol. 2002) ovlivňuje rychlost růstu a zpomaluje jej. Nicméně, nejvyšší tělesná hmotnost a hmotnostní přírůstek byly pozorovány v systémech individuálního ustájení (Xiccato a kol., 1999). Další studie autorů Matics a kol. (2018) zaznamenala vyšší přírůstek tělesné hmotnosti u králíků umístěných v kleci na rozdíl od králíků umístěných v boxech s výsledkem lepšího poměru konverze krmiva ve věku králíků od 7 do 9 týdnů. Naopak studie Princze a kol. (2009) neprokázala žádný vliv velikosti skupiny na hmotnostní přírůstek nebo na konečnou živou hmotnost králíků. Autoři však prokázali pokles denního přírůstku hmotnosti u králíků z větších skupin, přičemž snížení bylo mezi 1,0 a 9,3 g/den (Princz a kol., 2009; Combes a kol., 2010). Na základě uvedených studií nelze jednoznačně konstatovat, že vliv systému ustájení na hmotnost před porážkou existuje. Na vzniklých rozdílech se patrně podílí více faktorů, ať už chování zvířat a s tím spojené agresivní výpady, nebo obecně interakce mezi zvířaty, které je stimulují k pohybu, tak prostor k pohybu samotný. Například v přiložených studiích Krunt a kol. (2021; 2022) je vliv systému ustájení

na parametry výkrmu nejednotný. V první zmiňované studii byl vliv ustájení zjištěn jako signifikantní a zvířata se tak v 80 dnech věku v porážkové hmotnosti lišila o 224 g, kdy králíci z klecí měli hmotnost vyšší než králíci z boxů. Naproti tomu, nebyly, v pořadí druhé citované studii, mezi králíky v klecích a boxech nalezeny statisticky významné rozdíly.

Hustota osazení

Dalším faktorem, který ovlivňuje produkční parametry, je hustota osazení ustájovacího prostoru. Když byla hustota osazení snížena z 20-23 na 15-16 králíků/m^2, parametry růstu se zlepšily (Morisse a Maurice, 1997). Mousa-Balabel (2009) a El-Bayoumi a kol. (2018) pozorovali nejnižší přírůstek tělesné hmotnosti u králíků chovaných při hustotě osazení 28 králíků/m^2 v porovnání s králíky chovanými při hustotě osazení 20 nebo 12 zvířat/m^2. Princz a kol. (2008a), Szendrő a kol. (2009), Szendrő a Dalle Zotte (2011) a Paci a kol. (2013) však nezjistili žádný efekt snížení hustoty osazení na nižší počet než 15-17 králíků/m^2. Nicméně, vyšší hustota osazení způsobila nižší příjem krmiva v období výkrmu dle studií autorů Moriss a Maurice (1997) a Trocino a kol. (2004).

Podlahy

V průběhu let byl zkoumán vliv různých typů podlah na produkční a jateční parametry u vykrmovaných králíků. Při porovnání tří různých typů podlah (kovový rošt x plastový rošt x hluboká podestýlka) byla živá hmotnost vyšší u králíků od 7 do 10 týdnů věku, chovaných na podlaze s plastovým roštem než na kovovém roštu a hluboké podestýlce. Rozdíl byl také u králíků ve věku 11 týdnů chovaných na plastových roštech a hluboké podestýlce. Z hlediska růstu byly zjištěny vyšší hodnoty živé hmotnosti ve prospěch králíků ustájených na plastových roštech v porovnání s králíky z hluboké podestýlky (Gerencsér a kol., 2014). Naproti tomu, Dalle Zotte a kol. (2009) porovnávali králíky vykrmované na kovových a plastových roštech s výsledkem statisticky nevýznamných rozdílů v růstu. Dal Bosco a kol. (2015) pak také neobjevili signifikantní rozdíly pro charakteristiky růstu u králíků vykrmovaných na kovových a plastových roštech. Naopak, Trocino a kol. (2015) zkoumali rozdíl mezi plastovým a dřevěným roštem s výsledkem vyššího denního přírůstku hmotnosti, příjmu krmiva a živé hmotnosti ve prospěch plastových roštových podlah oproti dřevěné roštové podlaze. Dále pak bylo používání hluboké podestýlky, jakožto prvku zakrývajícího podlahu, zhodnoceno jako nevhodné z produkčního hlediska. Hluboká podestýlka totiž způsobila nižší přírůstky živé

hmotnosti, díky její konzumaci králíky, v důsledku čehož byl omezen příjem granulí (Lambertini a kol., 2001; Matics a kol., 2014).

1.3 Vliv systému ustájení na parametry jatečného těla

Velikost skupiny

Vliv velikosti skupiny na znaky jatečně upraveného těla byl potvrzen několika autory (Dal Bosco a kol., 2002; Dalle Zotte a kol., 2009; Combes a kol., 2010; Matics a kol., 2014). U králíků umístěných ve větších skupinách byla zjištěna nižší porážková hmotnost, jatečná výtěžnost a vyšší vývoj zadních partií v porovnání s králíky z menších skupin. To je dle autorů způsobeno vyšší fyzickou aktivitou zvířat v takovýchto skupinách (Combes a kol., 2010). Dle těchto autorů je pak takovým pohybem sníženo procento tuku v těle takto vykrmovaných zvířat a snižuje se poměr masa ku kosti. Loponte a kol. (2018) uvádějí, že u králíků chovaných ve volných systémech ustájení byla pozorováno nízké procentuální zastoupení tuku v jatečně upravených tělech než u králíků chovaných v kleci. To autoři vysvětlili vyšším výdejem energie při pohybu, skákání a běhu. Některé kontroverzní výsledky publikovali Machado a kol. (2019), kteří nepozorovali žádný vliv systému ustájení (klece vs. box) a velikosti skupiny (3 vs. 6 králíků na systém ustájení) na jatečnou výtěžnost, procento tuku a vývin zadních končetin. Autoři uvedli, že se zvířata na daný systém a management chovu v průběhu času adaptovala, což vedlo ke statisticky nevýznamným rozdílům. Na druhou stranu, Metzger a kol. (2003) zjistili statisticky vyšší porážkovou hmotnost a jatečnou výtěžnost ve prospěch králíků chovaných v boxech v porovnání s těmi v klecích a Matics a kol. (2018) uvedli vyšší vývin zadní části u králíků chovaných v boxu v porovnání s těmi v kleci. Nicméně, ve studii Krunt a kol. (2022), mimo jiné, zjistili, že se systém ustájení významně podílí na vývinu různých svalů. Obecně známý je právě rozdílný stupeň vývinu svaloviny stehna králíků, kteří jsou vykrmováni odlišně (zpravidla klec vs. výběh/box). Většina studií ale ve svých výsledcích zhodnocuje pouze největší sval stehna – *biceps femoris*. Ve výše zmiňované studii byl ale také zhodnocen například *quadriceps femoris*, pro nějž data ve vědecké literatuře, v kontextu s ustájením u králíků, chybí.

Hustota osazení

Dalším faktorem ovlivňujícím parametry jatečného těla je hustota osazení. Pokud byla hustota osazení vyšší než 15-17 králíků/m^2, způsobila vyšší jatečnou výtěžnost (Trocino a kol., 2004). Snížením hustoty osazení z 16 na 12 králíků na klec se výrazně zvýšila hmotnost jatečně

upraveného těla (Trocino a kol., 2015). Nejlepší parametry jatečných těl byly zjištěny při hustotě osazení 5 králíků/m^2. Ve studiích Dal Bosco a kol. (2000) a Pla (2008) bylo zaznamenáno, že obecně při zvýšení hustoty osazení systému ustájení králíky klesá podíl zadních končetin. To je v souladu se zjištěními autorů Matics a kol. (2018) o vyšším vývoji zadní části, který je více upřednostňován spotřebiteli (Dal Bosco a kol., 2002).

Podlahy

Vliv typu podlahy na parametry jatečně upraveného těla nebyl podle Princze a kol. (2009) signifikatní. Na druhou stranu, Trocino a kol. (2015) zjistili vyšší živou a jatečnou hmotnost a jatečnou výtěžnost u králíků chovaných na plastové roštové podlaze než na dřevěné podlaze. Tito králíci měli také vyšší poměr svalů ke kostem v zadní části jatečně opracovaného trupu. Jatečná výtěžnost byla dle studie Trocino a kol. (2008) výrazně vyšší na drátěné podlaze ve srovnání s podlahou z pokovovaných lišt, plastových lišt, a slámou na drátěné podlaze. Nicméně, Dal Bosco a kol. (2002) ve své studii konstatovali, že když je růst výrazně snížen kvůli nevhodné podlaze, jsou narušeny i znaky kvality jatečně upraveného těla a masa.

1.4 Vliv systému ustájení na kvalitu masa – fyzikální a chemické parametry

Králičí maso je bohatým zdrojem bílkovin a esenciálních aminokyselin a má vysokou nutriční hodnotu. Nejčastějšími kyselinami v králičím mase jsou nasycené mastné kyseliny (SFA) a polynenasycené mastné kyseliny (PUFA). Kvalita masa z pohledu zdravotního stavu závisí na obsahu SFA a tuku. V této části budou shrnuty účinky systémů ustájení na složení masa (Szendrő a Dalle Zotte, 2011).

Velikost skupiny

Vliv velikosti skupiny na fyzikální vlastnosti masa, jako je pHu (24 h postmortem) nebo barva masa byl zjištěn několika kolektivy autorů (Dal Bosco a kol., 2002; Dalle Zotte a kol., 2009; Combes a kol., 2010; Xiccato a kol., 2013; Matics a kol., 2018), kteří sledovali vliv různé úrovně stresu na barvu masa. Agresivní chování ve větších skupinách a související stres u králíků umístěných v boxech vedly k reakci svalů, které změnily barvu a měly nižší hodnoty pHu v porovnání se zvířaty z menších skupin (Matics a kol., 2018). Naopak, Lambertini a kol., (2001) nezjistili vliv velikosti skupiny na barvu masa. Co se týče hodnot pHu, ty byly vyšší u svalů hřbetu u králíků chovaných v kleci v porovnání s těmi v boxech (Dal Bosco a kol., 2002; Dalle Zotte a kol., 2009).

Combes a kol. (2010), Xiccato a kol. (2013) a Palka a kol. (2018) však nenalezli žádné změny pHu mezi jednotlivými systémy ustájení, co se jednotlivých jatečných partií týče. Naproti tomu, Lazzaroni a kol. (2009) pozorovali vyšší pHu u *biceps femoris* a *longissimus lumborum* u králíků umístěných v boxech v porovnání s těmi v klecích. Autoři se ale domnívají, že tomu tak bylo v důsledku odchytu králíků před porážkou. Szendrő a Dalle Zotte (2011) uvedli vliv velikosti skupiny na hodnoty červenosti (a*) a žlutosti (b*) jako nejasný. Dal Bosco a kol. (2002) a Dalle Zotte a kol. (2009) zjistili, že hodnoty barvy L (světlost)*a*b* byly vyšší u králíků chovaných v kleci v porovnání s těmi v boxech. Naproti tomu Combes a kol. (2010) a Mattioli a kol. (2016) zaznamenali tyto hodnoty nižší u králíků chovaných v klecích oproti těm v boxech. Szendrő a Dalle Zotte (2011) nabídli vysvětlení, že hodnota L* se nezmění, pokud pH není ovlivněno systémem ustájení (velikostí skupiny, respektive). Dal Bosco a kol. (2002) uvedli, že když se zvýší množství SFA a mononenasycených mastných kyselin (MUFA) v tuku, zvýší se i obsah PUFA. Szendrő a Dalle Zotte (2011) vysvětlili tento fenomén jako efekt ustájení větších skupin králíků s klesajícím obsahem tuku v mase, kdy odezvou bude zvýšení relativní koncentrace PUFA v mase.

Hustota osazení

Podle autorů Szendrő a Dalle Zotte (2011) není vliv hustoty osazení na složení masa tak jasný, jak by měl být, protože se zkoumaly jen hustoty osazení nižší než 17 králíků/m^2. Preziuso a kol. (2009) zjistili vyšší hodnoty pro a* a nižší pro L* u králíků chovaných při hustotě osazení <5 králíků na m^2. Později, Matics a kol. (2014) porovnali hustoty osazení 10,5 králíků/m^2 a 16,3 králíků/m^2 bez významných rozdílů pro barvu u svalu *longissimus dorsi*. Naopak, Dalle Zotte a kol. (2009) a Paci a kol. (2013), zjistili vyšší hodnoty L* u masa hřbetu králíků při vyšší hustotě osazení oproti těm s nižší hustotu osazení. Vliv hustoty osazení (10 vs. 4 králíci/m^2) na celkový obsah PUFA a MUFA zjistili Volek a kol. (2014) ve prospěch vyšší hustoty osazení.

Podlaha

Profil mastných kyselin ve svalech podle autorů Dal Bosco a kol. (2015) může také ovlivnit typ podlahy. V této studii byly pozorovány rozdíly mezi kovovým roštem, plastovým roštem a hlubokou podestýlkou. Méně MUFA u *longissimus thoracis et lumborum* bylo zaznamenáno na podlaze z kovového roštu. PUFA byly na nejnižší úrovni na hluboké podestýlce. Tyto hodnoty lze diskutovat právě s podobnými studiemi, např. Dalle Zotte a kol. (2009) zjistili, že PUFA se nezměnily ve dvou různých typech ustájení (klec vs. kotec) bez

významného vlivu typu podlahy (kovový rošt vs. plastový rošt), na rozdíl od autorů Dal Bosco a kol. (2002), kteří zjistili významně vyšší hladinu PUFA u *longissimus lumborum* a Chodová a kol. (2014) uvedli vyšší obsah PUFA u králíků na slámě v boxu než u králíků v kleci na kovovém roštu.

1.5 Vliv systému ustájení na vývin svalových vláken

Z hlediska zastoupení Lefaucher (2010) uvádí, že se svaly skládají ze svalových vláken typu I (βR) a typu II (αR, αW). U králíků se většinou sledují dva nejvíce dominantní svaly (*biceps femoris* a *longissimus thoracis et lumborum*). Svalová vlákna ovlivňují vývoj posmrtných změn, a i kvalitu masa (Hernández a kol. 2006). Dvě základní charakteristiky, které definují svalová vlákna, jsou průměr a obvod. Třetím určujícím faktorem, který ovlivňuje velikost svalu, je plocha průřezu (Chodová a kol. 2014).

Hodnocení vlivu hustoty osazení na charakteristiky svalových vláken ve svalu *biceps femoris* realizovali Volek a kol. (2014). U hustoty osazení (10 králíků/m^2 vs. 4 králíci/m^2) byl podíl αW vláken 79,3 vs. 59,2 %. Při srovnání stejných hustot byl podíl vláken αR 24,5 vs. 14,2 %. Naopak, distribuce vláken βR byla vyšší při nižší hustotě osazení (16,3 %), zatímco při vyšší hustotě osazení pouze 6,5 %. Plocha svalových vláken typu βR byla významně nižší u králíků chovaných při nižší hustotě osazení v porovnání s těmi s vyšší hustotou osazení ustájovacího prostoru (1 882 vs. 2 744 μm^2), plocha vláken αR a αW nebyla signifikantně ovlivněna. Chodová a kol. (2014) diskutovali dva systémy ustájení (skupinová klec s kovovým roštem vs. box se slámou) a jejich vliv na vývoj svalových vláken u *biceps femoris*. Byla ale použita odlišná nomenklatura typizace svalových vláken než u dříve citovaného článku. Plocha průřezu vláken typu I byla významně větší u králíků chovaných v klecích s vyšší hustotou osazení a jejich průměr byl také větší ve srovnání s plochou průřezu a průměrem svalových vláken u králíků chovaných při nižších hustotách osazení.

V další studii (Krunt a kol., 2021) byl zkoumán vliv klecového a skupinového ustájení v boxech na vývin svalových vláken u *biceps femoris*, přičemž byly objeveny dvě tendence, které ukazovaly vyšší hodnotu obvodu a plochy průřezu svalových vláken typu βR u králíků vykrmovaných v boxu než u králíků v kleci.

1.6 Možnosti obohacení ustájení a jejich vliv na výkrm králíků

Ve vědeckých studiích bylo zkoumáno několik typů obohacení a jeho vliv na produkční charakteristiky, případně pak na charakteristiky jatečně upravených těl. Mezi ty patří například okus (Buijs a kol., 2011; Zucca a kol., 2012), zrcadla (Reddi a kol., 2011; Musco a kol., 2019) nebo poličky (Matics a kol., 2018).

Okus

Vliv okusu na užitkovost a znaky jatečně upraveného těla králíků pozorovali Mohammed a Nasr (2016). Výsledkem bylo, že králíci, kteří měli k dispozici okus, měli vyšší konečnou hmotnost při porážce, celkový přírůstek hmotnosti, denní příjem krmiva a vyšší hmotnost jatečně upraveného těla. Princz a kol. (2008a), Buijs a kol. (2011) a Zucca a kol. (2012) nezjistili žádný vliv okusu na užitkovost králíků.

Zrcadla

Dalším zkoumaným prvkem jsou zrcadla. Musco a kol. (2019) doporučují použití zrcadel ve volném výběhu králíků. Umístění zrcadel v tomto systému ustájení vedlo ke zlepšení vlastností jatečně upravených těl, zvýšení přírůstku hmotnosti a vyšší jatečné výtěžnosti. Králíci dle autorů soustředí svou energii na průzkum zrcadel, což snižuje jejich pohybovou aktivitu a zvyšuje charakteristiky růstu. V systému individuálního ustájení měla zrcadla vliv na konečnou živou hmotnost králíků (Reddi a kol., 2011), která byla vyšší u těch, jež zrcadlo k dispozici měli než u těch bez zrcadel.

Poličky

Poličky jsou obecně používaným typem obohacení a dnes jsou v rámci legislativy pro ČR nutností. Nicméně jejich účinek na produkční parametry je stále předmětem vědeckého výzkumu. Nepochybně se platformy používají k poskytování obohacení prostředí, zvýšení možnosti pohybu a k odstranění případného agonistického nebo stereotypního chování (Matics a kol., 2018). Až do dnešních dnů několik studií reportovalo výsledky bez významného statistického vlivu víceúrovňových poliček na růst králíků (Princz a kol., 2009; Matics a kol., 2018). Pouze lze konstatovat, že z hlediska hygieny, je třeba uvažovat vhodné umístění poličky v rámci daného systému ustájení (Trocino a kol., 2019).

1.7 Možnosti obohacení ustájení a jejich vliv na welfare králíků ve výkrmu

Snahy o intenzifikaci chovu králíků byly v průběhu let spojovány se systémem chovu a výběrem technologií, které by nejlépe splňovaly podmínky welfare a zároveň se vyhnuly omezením produkce v souvislosti s nevhodně zvoleným systémem ustájení nebo technologií chovu. Hlavní nedostatky z hlediska welfare je u intenzivně chovaných králíků nedostatek prostoru pro odpočinek, úkryt nebo přirozené pohyby, jako je poskakování a skákání (Verga, 2000). Newberry (1995) definoval obohacení jako zlepšení biologického fungování zvířat chovaných v péči člověka vyplývající z úprav jejich prostředí. Obohacení by mělo být použito

ke snížení negativních emočních stavů, jako je strach, stres spojený s vystavením novým podnětům, nuda a apatie z nevhodného ustájení. EFSA (2005) zdůraznila, že obohacení životního prostředí je důležité, aby zvířata měla příležitost vyjádřit své přirozené chování.

Typy podlah a poličky

Význam typu podlahy jako nejdůležitějšího technologického prvku a jeho dopad na pohodu zvířat byl částečně popsán v review autorů Szendrő a Dalle Zotte (2011). Typ podlahy hraje klíčovou roli, protože na ní zvířata tráví celou dobu a je jí ovlivněna jejich lokomoce a odpočinek. Existuje několik typů podlah, které lze použít pro vykrmované králíky s určitými výhodami i nevýhodami. Review je zaměřena na používání těchto typů podlah několika autory. Pokud je v následujícím textu uvedena v souvislosti s typem podlahy poznámka o lepším welfare, tak tuto podlahu králíci preferovali a jejich preference je reflektována jako součet času, který na ní strávili.

Trocino a kol. (2015) porovnávali dřevěné roštové podlahy a plastové roštové podlahy s lepšími výsledky welfare ve prospěch plastových roštových podlah. Gerencsér a kol. (2014) porovnávali kotce s podlahami z 1/3 řešené stejnou plochou drátěného roštu, plastového roštu a slaměné podestýlky, vše umístěné v jednom kotci. Lepší výsledky welfare zvířat byly zjištěny ve prospěch podlahy z umělé hmoty, kde králíci trávili nejvíce času. Dal Bosco a kol. (2015) porovnávali stejné typy podlah. Zjistili pouze negativní dopad používání hluboké podestýlky na zdravotní stav králíků. Dall Zote a kol. (2009) zjistili, že při srovnání typů podlah (drátěná rošt vs. plastový rošt), zvířata umístěná na plastových podlahách měla těžší přední části jatečně opracovaného trupu a hlavy, což mohlo být dle autorů způsobeno držením těla králíků, souvisejícím právě s již zmíněnou podlahou. Trocino a kol. (2018) použili dřevěné a plastové rošty. Podle jejich výsledků trávili králíci chovaní v kotcích s dřevěnými roštovými podlahami více času odpočíváním ve skrčené poloze než ostatní. Vykazovali také sníženou úroveň péče o srst, pohybu a okusování prvků systému ustájení.

Předmětem výzkumu vhodnosti materiálu, případně typu podlahy, bývá často také znečištění nohou králíků a jejich otlaky, protože podlaha hraje klíčovou roli v pohodlí a hygieně králíka (Masthoff a Hoy, 2019). Při porovnání tří typů podlah (plastová roštová podlaha s příčkami 5 mm, šířka drážky 13 mm, perforace 75 % x plastová roštová podlaha s příčkami 10 mm, šířka drážky 10 mm, perforace 50 % x plastová roštová podlaha s příčkami 10 mm , šířka drážky 10 mm, perforace 50 %, také v kombinaci s <15 % perforací podlahy na vyvýšené poličce x plastová roštová podlaha s příčkami 12 mm, šířka drážky 12 mm, 50% perforace na

podlaze a vyvýšené plošině), bylo znečištění zadních nohou na nejnižší úrovni u králíků chovaných na plastové roštové podlaze s 5 mm příčkami, šířkou drážky 13 mm a se 75% perforací. Další tři typy podlah vykazovaly výskyt otlaků, které byly způsobeny kontaminací podlahy.

Poličky jsou komerčně využívány na farmách v případě potřeby zvětšení plochy ustájovacího prostoru. Používání několika typů poliček, s hlubokou podestýlkou, drátěná síť bez trusníku nebo s trusníkem, bylo běžnou praxí v experimentální sféře i na farmách (Szendrő a kol., 2012). Při porovnávání různých typů plošin byla pro králíky vhodnější plastová roštová polička než polička z drátěného pletiva (Szendrő a kol., 2012; Gerencsér a kol., 2014). Použití poliček zvyšuje plochu systému ustájení a poskytuje králíkům lepší možnost pohybových aktivit. Králíci také preferovali poličky, nad nimiž byl jako vršek klece použit neperforovaný materiál, protože se pod ním snažili schovat (Matics a kol., 2018). Szendrő a kol. (2012) toto chování vysvětlili tím, že evropští divocí králíci si přirozeně vyhrabávají nory pod zemí, aby našli úkryt a tím pádem se tento rys chování vyskytuje i u domestikovaných králíků, kteří mají tendenci hledat bezpečný prostor pro schování se. Ve studii Maticse a kol. (2018) byla řešena problematika možného vyššího indexu morbidity a mortality u králíků ustájených ve větších skupinách s využitím poliček. Problém spatřovali autoři v používání roštových platforem nebo mezipater, které na jedné straně zlepšují pohodu zvířat, umožňují králíkům větší pohyb a snižují stres, ale naopak králíci se pod nimi spíše schovávají. Skutečným problémem králíků skrývajících se pod plošinami, které jsou umístěny v rozích boxů, je pak situace, kdy na ně králíci nad nimi močí a kálí. Olizování (druhově specifické chování králíka) podlahy je pak hlavním faktorem, který vede k zvýšenému infekčnímu tlaku působícího na mladé vykrmované králíky. Možným východiskem by tedy mohla být implementace těchto plošin do středu boxů, což králíkům zabrání v pocitu, že plošina je ideálním místem pro úkryt.

Okus

Využití okusu, jakožto obohacujícího prvku, má v intenzivních chovech zvířat velký význam. Jeho využití je známo například u lišek, které si prostřednictvím okusování těchto předmětů uspokojují tuto svou druhově specifickou potřebu (Lapinski a kol., 2019).

U myší je toto obohacení instalováno, aby se zabránilo nadměrnému růstu zubů (Henkel a kol., 2018) a také u kupírovaných prasat se používá něco podobného jako klacky (literatura uvádí pouze označení jako dřevěné laťky), aby se eliminovalo kousání ocasu jako negativní chování (Chou a kol., 2018). U králíků je k dispozici dřevěný okus k odstranění stereotypního

chování (Trocino a kol., 2013), jako je kousání nebo olizování mříží v kleci nebo nadměrné ošetřování srsti, které je ve zvýšené míře nežádoucí. Využití okusu králíky také snižuje hladinu kortizolu a potenciálně zlepšuje jejich životní pohodu (Mohammed a Nasr, 2017) a stimuluje králičí pozornost na jiné předměty s možným vlivem na relativní hmotnost mozku (Bozicovich a kol., 2016).

Ve vědecké literatuře bylo porovnáno několik typů materiálů, ze kterých je okus vyroben (Princz a kol., 2007; Mohammed a Nasr, 2017). Princz a kol. (2007) zjistili, že nejvyšší preference byla u lípy malolisté. Velmi konzumovány byly také jírovec bílý a vrba bílá. Preference vrby potvrdili i Mohammed a Nasr (2017). Výše uvedené materiály byly oblíbené pro svou tvrdost, vůni a chuť. Autoři považovali za nevhodné pro použití jako obohacení tyto druhy okusu: černý bez, modřín evropský, bříza bělokorá, moruše bělokorá a dub obecný. Princz a kol. (2008a) umístili okus (trnovník akát) jen do některých klecí, což vedlo (s možností preference klece s nebo bez okusu) k výběru obohacených klecí. Obecně lze říci, že na základě vědeckých poznatků, králíci preferují ve svých systémech ustájení přítomnost okusu (Princz a kol., 2008b; Buijs a kol., 2011; Bozicovich a kol., 2016). Nicméně, okus měl vliv i na chování zvířat. Sebe pečování a sociální kontakty, stres a agresivní chování které by za podmínek ustájení bez obohacení mohly vést ke zraněním, byly sníženy s vysvětlením interakce s obohacením (Verga a kol., 2004; Buijs a kol., 2011; Bozicovich a kol., 2016).

Umístění (podlaha vs. pod stropem) okusu je také důležité ve vztahu k poměru bakterií v systému ustájení (*E. coli*, *Clostridium*, *Lawsonia intracelularis*). Okusy by mohly být v technologiích farem kontaminovány králíky a mohlo by být ohroženo zdraví zvířat. Maríno a kol. (2018) pozorovali, že pokud měli králíci možnost vybrat si, kde budou okusovat dřevěný okus, činili to ve vyšším poměru na podlaze systému ustájení než pod stropem. Ačkoliv králíci preferují umístění okusu na podlaze, je tam jeho umístění nevhodné z důvodu mikrobiálního znečištění.

Zrcadla

Individuální ustájení králíků je typické především pro dospělé samice, které ve skupinovém ustájení mnohdy projevují zvýšenou míru agresivního chování (Edgar a Seaman, 2010) a pro samce. Skupinové ustájení samic je však v současnosti intenzivně zkoumáno z důvodu výše zmíněné agresivity, zranění mláďat a samotných samic, což do značné míry zpochybňuje úroveň welfare v těchto systémech ustájení (Szendrő a kol., 2019). Chu a kol. (2004) zjistili, že králíci chovaní jednotlivě vykazovali více stereotypního chování než králíci

chovaní v páru, pravděpodobně proto, že se jedná o společenská zvířata. K eliminaci stereotypního chování se zdá být vhodnou cestou využití zrcadel. Aby bylo možné porozumět tomu, jak reagují rostoucí králíci na zrcadla, je nutné porozumět dospělým králíkům. Edgar a Seaman (2010) popsali různé behaviorální reakce na podněty ve vztahu k pohlaví dospělých králíků. U samostatně ustájených králíků může péče o srst přejít ve žvýkání srsti nebo její tahání, případně vytrhávání. Autoři zjistili, že zrcadla způsobují u samic snížení péče o srst, což by mohlo naznačovat odstranění sociální deprivace, protože právě zvýšená péče o srst naznačuje opak. Na druhé straně se po aplikaci zrcadel do klecí samců zvýšila jejich ostražitost. Schofield (2019) vysvětlila tyto poznatky tím, že se králíci v zrcadlech vidí, obrazy rozeznávají jako specifické, a proto mohou u samců vyvolávat ostražitost s ohledem na sexuální chování. Samci také snížili péči o srst možná proto, že projevili teritoriální chování a soustředili tak většinu pozornosti k odrazu v zrcadle. V této situaci by zrcadla mohla být pro samce škodlivá, zejména na malých plochách.

U rostoucích králíků Jones a Phillips (2005) doporučovali zrcadla, protože stimulovala potřebu králíků zkoumat jejich prostředí (hrabání a čichání). Také toto obohacení může ovlivnit čas, který králíci tráví civěním z klece, aby ho nahradili zájmem o zrcadla. Preference části systému ustájení se zrcadly zjistili i Dalle Zotte a kol. (2009). Zjistili také nejvyšší preferenci částí kotce, kde byla umístěna zrcadla, u králíků ve věku 5,5 – 8,5 týdne, což ukázalo, že králíci se raději shlukovali do větších skupin, než aby byli o samotě. Vysoký zájem o zrcadla u mladých králíků popsali Edgar a Seaman (2010). Konstatují, že králíci si na nový objekt postupně zvykali a považovali ho za méně zajímavý na konci výkrmu než v první části výkrmového období, kdy už byla míra zájmu nižší. Mastellone a kol. (2019) a Musco a kol. (2019) doporučovali použití zrcadel při chovu králíků ve volném výběhu. Králíci mohli soustředit svou energii na zkoumání zrcadel, což snížilo jejich pohybovou aktivitu (a zlepšilo přírůstek živé hmotnosti) a také neprojevovali negativní chování ve smyslu agresivity nebo stereotypů. Na druhou stranu, Schofield (2019) poukázal na to, že přítomnost zrcadla by mohla vést ke zvýšení příjmu krmiva (a následně i hmotnosti) z důvodu projevu konkurenčního chování, hlavně v pozdní fázi výkrmu.

1.8 Stres a kvalita kostí jako indikátor welfare

V životě zvířete existují různé strategie chování a fyziologické odpovědi, které reagují na stresory. Jedná se o stimulaci sympatického nervového systému, která vede k uvolnění katecholaminů (adrenalin, noradrenalin), a aktivaci osy hypotalamus-hypofýza-nadledviny (HPA). HPA má důležitou roli. Obnovuje homeostázu, což má za následek sekreci

glukokortikoidů (kortizol, kortikosteron). Tento stav může trvat minuty až hodiny (Sheriff a kol., 2011). Když jsou zvířata vystavena krátkodobým problémům, bolestivým stavům, jsou manipulována, přepravována nebo jsou konfrontována s neobvyklou situací, hladina kortizolu se zvyšuje. Toto zvýšení hladiny kortizolu může být indikátorem špatného welfare (Broom, 2017). Krátkodobé situace vedou ke krátkodobému zvýšení koncentrace glukokortikoidů, které pomáhají zvířeti uniknout. Nicméně škodlivé následky jsou způsobeny ve spojení s kondicí zvířete (Blas a kol., 2007).

Kortikosteron a agresivní chování

Glukokortikoidy jsou obecně nástrojem pro pochopení toho, jak stresory a přírodní prostředí (predátoři, počasí), změna klimatu nebo přemístění atd. ovlivňují populace (Monclús a kol., 2009). Existuje několik typů měření těchto hormonů; krev (plazma nebo sérum), sliny, moč, výkaly, vlasy a peří (Sheriff a kol., 2011). U králíků jsou typickými vzorky pro měření výkaly a jsou nejčastěji používaným médiem pro hodnocení stresu (Trocino a kol., 2014; Matics a kol., 2018). Vzorkování chlupů není tak rozšířené, jak by mělo být (Comin a kol., 2012). Chlupy jsou však poměrně stabilní, jejich odběr je neinvazivní (Balíková, 2005) a analýza nabízí hodnocení chronické aktivity osy HPA (Peric a kol., 2017). Skutečným problémem v ustájení králíků je nejasný názor na podmínky ustájení, obohacování a ustájení králíků podle pohlaví. Tyto faktory mohou vyvolat strach, negativní emoce, což má za následek negativně ovlivňující welfare králíků a celé období výkrmu (Peric a kol., 2017).

Několik studií zkoumalo vliv podmínek ustájení na pohodu králíků (Trocino a kol., 2014; Matics a kol., 2018). Indikátorem byl stresový hormon – kortikosteron. Tento hormon má stejnou funkci jako kortizol (Broom, 2017), takže odkaz na kortizol v níže uvedeném textu se vztahuje na kortikosteron. Szendrő a Dalle Zotte (2011) poukázali na vyšší míru agresivity a vzájemného napadání králíků, kteří byli umístěni ve skupinových boxech. Jejich tvrzení je podloženo studií autorů Maertense a Van Herck (2000), kteří se domnívají, že vysoký stupeň agresivity králíků umístěných v boxech nebo klecích při vysoké hustotě osazení (obvykle v boxech nebo zvětšených klecích) způsobuje časté útoky na slabší nebo méně dominantní jednotlivce. Tato opatření by také mohla snížit příjem krmiva a narušit dobré životní podmínky zvířat v souvislosti s nemožností přijímat krmivo. Incidenci útoků hodnotili Rommers a Meijerhof (1998), kteří připustili možnost ovlivnění věku a hustoty osazení. Závěrem lze říct, že více kožních lézí spojených s vyšší frekvencí napadání se, bylo u králíků připisováno spíše hustotě osazení než věku.

Na začátek tohoto odstavce je třeba konstatovat, že abnormální chování, jako jsou stereotypy nebo vysoká míra agrese, jsou někdy velmi užitečným indikátorem špatného welfare v situacích, kdy nedochází ke zvýšené hladině kortizolu. Kolísání kortizolu nemusí být důkazem špatného welfare, protože je přípravou na námluvy nebo aktivní krmení (Broom, 2017). Proto kontext, ve kterém dochází ke změnám kortizolu, je zásadní informací pro interpretaci fyziologických dat. V nedávné studii (Matics a kol., 2018) byla zjištěna vyšší hladina kortikosteronu v trusu králíků umístěných v kotcích ve srovnání s králíky v kleci. Hladiny kortikosteronu se zvyšovaly s věkem králíků v boxech. Mezi šestým a jedenáctým týdnem se jejich hladina zvýšila téměř o 5procentních bodů. Také Trocino a kol. (2014) zjistili významně vyšší hladiny kortikosteronu u králíků umístěných v boxu ve srovnání s králíky v klecích. Jejich studie však byla založena na měření hormonů z chlupů. Dále, kortikosteron ve výkalech zkoumali Cornale a kol. (2016), kteří zaznamenali zvýšení hladiny kortikosteronu se snížením dostupného prostoru v klecích. Na druhou stranu, Buijs a kol. (2011) nepozorovali vyšší distribuci glukokortikoidních metabolitů ve výkalech v závislosti na velikosti klece. V jejich studii bylo stanoveno doporučení eliminovat strach jako psychický stav stresu ze změny velikosti skupiny nebo po naskladnění. Doporučili klec obohatit o předmět, který by simuloval možnost jednoduše dimenzovaného úkrytu ze tří dřevěných desek tvarovaných do písmene „U“. Trocino a kol. (2014) uvedli, že eliminace stresu a agonistického chování u rostoucích králíků je možná obohacením systému ustájení již dříve zmíněného okusu.

Dalším faktorem, který ovlivňuje welfare (případně vyvolává stavy strachu a agresivity), může být složení skupiny podle pohlaví (Bozicovich a kol. 2016). Jejich studie dospěla k závěru, že výkrm králíků podle pohlaví může způsobit méně zranění. Zjistili, že nejnižší počet zranění byl pozorován ve skupinách samic a nejvíce zraněných králíků bylo ve skupinách samců. Naproti tomu, Szendrő a kol. (2012) uvedli, že jako první útočí samice ve věku sedm, osm a devět týdnů. U králíků starších deseti a jedenácti týdnů věku byla frekvence poranění častější ve skupinách samců. Zranění způsobená agonistickými reakcemi měla vyšší frekvenci ve skupinách jedinců stejného pohlaví než ve smíšených skupinách. Králíci nejvíce útočili, když umělé světlo bylo podobné soumraku a úsvitu, jako v přírodních podmínkách. To odpovídá jejich druhově specifickému chování, kdy jsou králíci nejaktivnější za soumraku nebo za úsvitu. Nicméně Vervaecke a kol. (2010) zjistili, že agresivní interakce mezi zvířaty nejsou podle nich zaměřeny cíleně na stejné pohlaví.

Kvalita kostí ve spojitosti s pohodou zvířat

V období výkrmu nebývá se zlomeninami kostí problém, ale v případě manipulace se zvířaty je výskyt zlomenin častější (Martrenchar a kol., 2001). Co se zdravotního stavu kostí týče, největším přínosem u králíků vykrmovaných ve skupinových kotcích, je zvýšená odolnost proti zlomenině stehenní a lýtkové kosti, v porovnání s králíky ustájenými v klecích (Xiccato a kol., 2013). Dalle Zotte a kol. (2009) také potvrdili, že zlomeniny stehenní a holenní kosti jsou častější u králíků v kleci a dodali, že obě kosti mají vyšší hmotnost u králíků umístěných ve skupinách v boxech. Combes a kol. (2010) zjistili, že králíci v boxech měli vyvinutější zadní část jatečně opracovaného trupu než králíci v kleci, což indikovalo vyšší osvalení a vyšší hmotnost této části trupu. Autoři uvedli, že četnost zlomenin kostí je vyšší u králíků v kleci, zatímco králíci v boxech vykazují vyšší odolnost vůči zlomeninám. Výsledky těchto studií podporují tvrzení dřívější studie Dalle Zotte a kol. (2009), kteří uvedli, že snížení frekvence výskytu zlomenin lze docílit zvýšením plochy klece. Matics a kol. (2018) také zjistili, že králíci z boxu mají hmotnost kosti v porovnání s králíky z klecí nejen vyšší, ale tyto kosti jsou u nich zároveň delší. Krunt a kol. (2021) také porovnali kvalitu kostí králíků vykrmovaných v klecích s těmi vykrmovanými ve skupinových boxech a zjistili, že králíci ustájení v boxech měli vyšší pevnost lýtkové kosti, a také vyšší zastoupení popelovin, vápníku a hořčíku. V pevnosti stehenní kosti se králíci z výše jmenovaných systémů ustájení statisticky nelišili, ale obsah popelovin a hořčíku u těchto kostí byl signifikantně vyšší u králíků v boxech.

1.9 Ustájení chovných samic z pohledu welfare a zdraví

Ustájení chovných samic králíků je dlouhodobě diskutované téma napříč vědeckou i chovatelskou veřejností. V posledních letech zde byl vyvíjen relativně velký tlak na rozvolnění stávajících systémů, což byly individuální klece, na systémy skupinové – různými způsoby modifikované. Iniciace této změny prakticky pochází z teze, která říká, že je králík ve volné přírodě skupinově žijící zvíře. Aspekty života králíka v přirozeném prostředí jsou ale složitější a mnohem více komplexní, než kdy život zvířat v umělém ustájení bude.

Evropský králík divoký (*Oryctolagus cuniculus*) žije v koloniích, které mají přísně nastavenou hierarchii, které je zpravidla dosahováno silným teritoriálním chováním zvířat, které je doprovázeno různě intenzivními boji nebo kontakty. Skupina králíků žijící v kolonii se většinou skládá ze dvou až tří samců a dvou až devíti samic. Hierarchie je známa mezi oběma pohlavími, kdy samice bojují o místa k budování nory čili o místa k porodu a samci bojují o postavení ve skupině, s čímž souvisí přednostní právo k páření (Bell, 1983). Postavení v sociálním žebříčku se s věkem zvířat mění, a to s ohledem na jejich kondici. Boje mezi

samicemi jsou nejčetnější na začátku rozmnožovací sezóny, případně, jsou-li do skupiny zařazeny nové mladé samice. Poté se hierarchie ve skupině stabilizuje a frekvence bojů se sníží (Mykytowycz, 1958). Vytvoření jakýchsi skupin či částečných kolonií bylo cílem vědců posledních dekád. Agresivita mezi samicemi a vzájemné útoky v omezeném prostoru uměle vytvořených systémů ustájení ale do značné míry znemožňují snahy pro uspokojivé výsledky v oblasti produkce těchto králíků. Proto je stále většina samic na farmách ustájena individuálně se svými králíčaty až do odstavu (Mondin a kol., 2021).

Klecové systémy

Samice v reprodukci jsou na komerčních farmách chovány převážně v individuálních klecích, a to se svými mláďaty až do odstavu. Ve většině případů se jedná o klece s dvojím využitím, tzv. „dual purpose cages“, které umožňují samici okocení, odkojení mláďat a její setrvání s odchovem v jedné ubikaci až do odstavu. Odstavená králíčata pak v kleci zůstávají, odebrána je pouze samice, v tu dobu zpravidla znovu březí. Oproti předešlým letům jsou tyto klece vybaveny poličkou pro únik samice před mláďaty, případně pro její odpočinek a později i odpočinek mláďat. Navíc jsou v klecích k dispozici většinou i prvky určené k okusu a částečnému naplnění potřeb králíka ohlodávat předměty a brousit si zuby (Szendrő a kol., 2019).

Pokud jde o velikost klecí pro reprodukci samic, jejich rozměry mohou ovlivnit pohodu zvířat z hlediska možnosti poskytování vhodného prostoru pro pohyb a odpočinek. Délka a šířka především určují dostupnou plochu pro pohyb a odpočinek králíků, zatímco výška může ovlivnit obecné možnosti pohybu a některé konkrétní chování králíků, jako je například panáčkování. Omezení plochy pro pohyb může způsobit psychický stres, jako je nuda, frustrace a stereotypní chování (Verga a kol., 2007), stejně jako vývojové a kostní abnormality. Afektivní stavy spojené s nudou jsou pak častými iniciátory agresivních útoků u samic ve skupinách nebo důsledkem výskytu abnormalit jako je například vytrhávání srsti bez zjevné příčiny (příprava hnízda). Studie z minulého tisíciletí o ustájení samic v reprodukci ukázaly, že chov v malých a nízkých klecích souvisel s deformacemi páteře, a tedy špatnou úrovní welfare (Drescher, 1996). Podle autorů této studie, donutila nízká výška běžných klecí (<32 cm) zvířata přijmout prodlouženou polohu těla při sezení a pravděpodobně vyvolala systémovou hypoplazii kostní tkáně a také kaudální dislokaci těžiště těla v důsledku hmotnosti dělohy s plody. Buijs a kol. (2015) ale nenašli žádný vliv rozměrů klece na výskyt deformací páteře u samic chovaných v systémech s malým/nebo bez omezení výšky, tedy v individuálních klecích (3 952 cm^2/samice,

maximální výška stropu 63 cm) vs. systému pro poloskupinový chov (5 000 cm^2/samice, bez stropu). Negretti a kol. (2010) pozorovali, že nejběžnější postoje, které dospělí králíci zaujímají, nevyžadují víc než 40 cm výšky: zvířata stála pod touto výškou v 99,5 % pozorování (45 000 záznamů na 10 zvířatech během 1 týdne). Autoři uvedli, že standardně králíci nepanáčkují, tedy nestaví se na zadní nohy, pokud to nevyžaduje situace. Běžně se totiž jedná o průzkumné nebo ostražité chování. Králíci provádějí vzpínání nebo také natahování se po stěnách kotce, případně panáčkují, když jsou umístěni v novém prostředí, jako například při přesunu z běžné klece do do jiného ustájení (Olivas a kol., 2013). Motivace k postavení se na zadní nohy za podmínek chovu může souviset s ostražitým chováním v reakci na vizuální podnět (např. lidskou přítomnost) nebo hluk, který je v blízkosti chovného zařízení.

V individuálních klecích samice tráví většinu času v klidu (Alfonso - Carrillo a kol., 2014) a pohybují se velmi omezeně. Bylo také pozorováno, že mladší samice jsou ve svém ustájení aktivnější než samice starší (Bignon a kol., 2012). Na druhou stranu, u samic v reprodukci, které byly chovány ve standardních klecích ve srovnání s těmi, které byly chovány ve větších klecích (83 × 38 × 32 cm vs. 113 × 46 × 46 cm), byly naměřeny vyšší hladiny kortikosteronu ve výkalech (Prola a kol., 2013). Na výši hladiny kortikosteronu ve výkalech má vliv více faktorů a v ustájení králíků, a jeho vlivu na hladinu tohoto hormonu, se výsledky studií poněkud rozcházejí.

Vybavení klecových systémů – podlaha

V minulosti byly testovány různé typy podlah, ať už ve výkrmu králíků, nebo u samic v reprodukci. Na farmách se ale většinou používaly drátěné podlahy, které byly nejčastěji voleny z důvodu vysokých hygienických standardů – propadávání výkalů, jednoduchá dezinfekce atd. Hlavním nedostatkem tohoto typu podlah byla vysoká incidence otlaků – lézí na spodní části chodidla samic, různé závažnosti (Szendrő a kol., 2019). Incidence otlaků je pro chov králíků fatální, otlaky jednak omezují pohyb samic, dále jsou velmi bolestivé a tím kompromitují welfare, a také jsou potenciálním místem vzniku ložiska infekce (Ruchti a kol., 2019). Výskyt lézí v oblasti spodní strany tlapky je běžným zdravotním problémem u králíků, zejména u těch chovaných jako domácí mazlíčci, u nichž jsou obvykle poranění kůže doprovázena sekundárními smíšenými infekcemi, které často končí vytvořením hnisavých abscesů a ulcerací. U králíků se prevalence lézí na spodní straně tlapky týká právě především chovných králíků, zejména samic, kvůli vyšší délce dožití a vyšší tělesné hmotnosti, ve srovnání s králíky ve výkrmu (Rosell a de la Fuente, 2013). Léze na spodní části končetin ale nevznikají

pouze z otlačení na drátěné podlaze. Další příčinou může být nedostatečná výměna podestýlky v kotcích s hlubokou podestýlkou (slámou). Dlouhodobé sezení zvířat ve vlhké podestýlce má pak stejný efekt jako zatěžování končetin na drátěné podlaze. I proto jsou v současnosti klece většinou vybaveny plastovými roštovými podlahami, které při řádné dezinfekci a zoohygieně snižují incidenci otlaků u samic v reprodukci. Existují i studie, ve kterých bylo zaznamenáno, že samice ustájené na plastové podlaze vykazovaly méně agresivního chování v porovnání se samicemi, jejichž ustájovací podlaha byla z kovu. Autoři této studie (Zomeño a kol., 2018) však tyto výsledky nedokázali vysvětlit.

Vybavení klecových systémů – obohacující prvky

Obohacující prvky mají primárně za cíl zvyšovat welfare zvířat a přibližovat jejich život v uměle vytvořeném systému ustájení co nejblíže skutečným potřebám. Jedním z těchto prvků jsou platformy, nebo také poličky, jejichž funkce byla popsána v kapitole o výkrmu králíků. Kromě toho, že poličky poskytují králíkům více prostoru, tedy zvyšují plochu kotce, mají pro samice v laktaci velmi specifickou funkci. Samice se na ně mohou schovat před mláďaty, která vylézají z hnízda a dožadují se krmení, což může být pro samici v uzavřeném prostoru stresující faktor. Později, když jsou mláďata dost stará, aby na poličku vyskočila, tráví samice většinu času pod ní (Mikó a kol., 2014). Z hlediska zdraví končetin je také vhodné umístit plastovou podložku (v případě, že je polička drátěná a má charakter roštu). Samice, které byly na druhém nebo v pořadí vyšším vrhu, tuto podložku využívaly, dle výsledků autorů Mikó a kol. (2014) častěji než samice na prvním vrhu. Pro samice v reprodukci byla také dříve testována místa, která sloužila k úniku nebo schování (Rommers a kol., 2014), jejichž potenciál však nebyl dle Huanga a kol. (2021) nikdy naplněn.

Dalším možným obohacením systému ustájení je okus. Okus může být ve formě větviček, klacíků, nebo slisovaných pelet. Například Maertens a kol. (2013) uvedli, že přítomnost okusu v podobě pelet nijak nezměnila četnost okusování konstrukce klece samicemi (okusování mříží klece patří k projevům stereotypního chování), ale naopak výrazně zvýšila jejich aktivní chování. To naznačuje možnost ovlivnění chování samic prostřednictvím těchto pelet, které se napříč vědeckými pracemi liší svým složením.

Skupinové systémy

Skupinový chov samic králíků v reprodukci je standardně realizován dvěma způsoby. Tím prvním je kontinuální systém a druhým je dočasný systém (Szendrő a kol., 2019). Kontinuální

systém chovu, kdy jsou samice ustájeny dohromady a zapuštěny buď samcem, nebo inseminovány, se zdál být nadějný, ale po letech výzkumu byl v podstatě zavržen. A to z důvodu vysoké míry agresivního chování samic při soubojích o místo k porodu. Agresivita často vyústí v poranění, která samicím způsobují problémy zdravotního charakteru, je narušen welfare a snižuje se produkce (Andrist a kol., 2013). V tomto typu ustájení také často dochází k vysoké incidenci falešné březosti (Mugnai a kol., 2009) nebo porodům více samic do jednoho hnízda (Hoy a Matics, 2016). Naproti tomu dočasné systémy, kde jsou samice ustájeny v rámci reprodukční periody dohromady jen určitý časový úsek, mnohdy dosahují podobných výsledků v reprodukci jako individuální systémy ustájení. Nicméně, úroveň produkčních parametrů mláďat těchto výsledků potom nedosahuje (Maertens a Debie, 2017). Příčinou je opět agresivní chování samic vůči ostatním samicím, mláďatům z cizích vrhů a boje v hnízdech samic (Buijs a kol., 2015).

2 Vědecké hypotézy a cíle práce

2.1 Vědecké hypotézy

Králíci ustájení na plastové roštové podlaze v boxech nebudou mít významně sníženou užitkovost a kvalitu jatečných partií oproti králíkům ustájeným na drátěné roštové podlaze v kleci při stejné hustotě osazení.

Králíci ustájení na plastové roštové podlaze v boxech budou vykazovat vyšší kvalitu kostí než králíci ustájení v klecích na drátěné roštové podlaze při stejné hustotě osazení.

Samice králíků ve výkrmu budou mít na konci výkrmu vyšší hmotnost než samci. Samice budou mít vyšší procentuální zastoupení αW svalových vláken než samci, zatímco samci budou vykazovat vyšší procentuální zastoupení αR a βR svalových vláken než samice při stejné hustotě osazení.

Zvýšená fyzická aktivita ovlivní velikost pohybových svalů a tím i jejich vlastnosti jako je výtěžnost, barva, nebo síla střihu. Tyto změny způsobí zvýšení obsahu bílkovin ve svalech, a zároveň dojde ke snížení obsahu intramuskulárního tuku a tím budou ovlivněny senzorické charakteristiky masa.

Samice králíků v reprodukci ustájené na podestýlce, které budou mít možnost hloubit své vlastní nory, budou vykazovat nižší míru agresivního chování vůči ostatním samicím, budou mít méně zranění na kůži, méně zraněných králíčat a tím pádem nižší úhyn králíčat v porovnání se samicemi ustájenými na podestýlce bez možnosti vytvářet vlastní nory.

Samice plemene meklenburský strakáč, které budou mít možnost hloubit své vlastní nory budou mít méně zranění na kůži, méně zraněných králíčat a tím pádem nižší úhyn králíčat v porovnání se samicemi masného hybridu Hyplus, který bude mít stejné podmínky ustájení.

Systém ustájení samic v reprodukci bude mít vliv na kvalitu kostí zvířat a jejich vybrané reprodukční ukazatele, ale neovlivní jejich zdravotní stav reflektovaný krevním profilem.

2.2 Cíle práce

Mezi cíle této práce patří posoudit:

- vliv systému ustájení na parametry užitkovosti a složení jatečně opracovaného trupu králíků
- vliv systému ustájení na vývin různých svalových partií a charakteristiky svalových vláken

- vliv systému ustájení na welfare prostřednictvím kvality kostí
- vliv pohlaví na produkční parametry, kvalitu jatečně upraveného trupu, vývin svalových vláken a kvalitu kostí
- vliv systému ustájení na fyzikální, chemické a senzorické charakteristiky kvality vybraných pohybových svalů zadní části králičího trupu spolu se svalem *longissimus thoracis et lumborum*
- vliv systému ustájení a genotypu na agresivní chování, incidence zranění a zdraví samic v reprodukci a jejich mláďat
- zdravotní stav a welfare samic v reprodukci prostřednictvím hematologických a biochemických ukazatelů krve, kvality kostí a jejich reprodukční ukazatele v různých systémech ustájení s/bez zrcadly/zrcadel

3 Seznam publikovaných prací

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A review of the effects of housing system on production and welfare in growing rabbits*

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The aim of this review was to evaluate the effect of different housing systems on productive traits, carcass, meat quality and muscle fibre properties in growing rabbits. Rabbit breeding for meat production is nowadays under the pressure of decreasing rabbit meat consumption and unsatisfactory animal welfare conditions. It is necessary to review which housing systems are the most suitable from the production point of view with respect to animal welfare. It is crucial to implement environmental enrichment of these systems in order to eliminate aggressive or stereotypical behaviour. There are several studies in scientific literature, which examined effects of group size, stocking density and floor types on productive traits, carcass traits, meat quality or welfare, but very few studies considered the potential impact of these factors on muscle fibre properties, which are the determining factor of carcass quality. Nowadays, more possibilities to enrich the housing system environment are available. Generally, gnawing sticks are used to eliminate the negative behaviour, while platforms have no effect on productive traits, although the exercise function is well received by rabbits. Additionally, mirrors may be used to decrease the effect of feeling isolated and thus improve welfare conditions.

KEYWORDS: carcass / housing systems / meat quality / productive traits / welfare

Housing of rabbits was confirmed as a factor, which influences productive performance [Dal Bosco *et al*. 2002, Dalle Zotte *et al*. 2009] and meat quality [Xiccato *et al*. 2013, Mattioli *et al*. 2016]. The effect of housing systems on muscle

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fibre characteristics is still in the research phase. Group size, stocking density and floor type were examined by several authors across the scientific spectre [Lambertini *et al*. 2001, Matics *et al*. 2014, Trocino *et al*. 2015]. Rabbits used for meat production conventionally are specifically hybridised strains and their meat is generally exported to most European markets [Cullere *et al*. 2018]. A commercial supply chain of this final product includes input suppliers, meat rabbit producers, abattoirs, logistic platforms, supermarkets and final consumers [Baviera-Puig *et al*. 2017]. Thus the best options need to be implemented for housing, stocking density, group size and other effects. EFSA [2020] provides a division of housing systems to meet the requirements of conventional farms and niche systems. The former group includes conventional cages, enriched cages and elevated pens. The latter group comprises floor pens, outdoor and organic systems. Individual housing systems have been used for a long time with the benefit of the best productive performance [Maertens and De Groote 1984] and superior meat quality [Xiccato *et al*. 2013]. Nevertheless, group housing systems with environmental enrichment have become increasingly popular because of the better welfare status of animals, which could have more social interactions with their mates [Buijs *et al*. 2011]. On the other hand, group housing of rabbits in groups bigger than 10 rabbits leads to a deterioration in productive performance, as well as lower carcass and meat quality [Dal Bosco *et al*. 2002, Xiccato *et al*. 2013]. The productive performance, carcass traits, meat quality and muscle fibres of growing rabbits in commercially used housing systems with their effects were compared in this study. The review is focused specifically on housing technologies, which according to scientific studies significantly influence previously mentioned parameters. These parameters (such as productive traits) are connected with animal welfare by affecting both animal well-being and performance thanks to environmental enrichment (e.g. gnawing sticks), with environmental enrichment being also a part of alternative housing systems.

Productive performance, carcass and meat quality characteristics

Productive traits

Growth, body weight gain or feed intake are these important characteristics, which have a serious impact on rabbit meat production. Several studies examined the effect of housing systems on productive performance with the proven greater growth and live weight in rabbits reared in cages compared to those of rabbits housed in pens of either small groups [Lambertini *et al.* 2001, Princz *et al*. 2009] or bigger groups [Dal Bosco *et al*. 2002, Combes *et al*. 2010]. The effect of group size, stocking density and floor type on productive performance will be discussed.

When comparing different group sizes [6, 12, 18, 30, 42, 54 individuals] no effect on growth and feed intake was recorded [Rommers and Meijerhof, 1998]. On the other hand, Xiccato *et al.* [1999] found that daily feed intake significantly decreased in animals kept in groups compared to individual cages. More free space was observed

in larger cages because animals tended to rest in one part of the cage. Locomotion activity increased with the greater available space and it negatively influenced feed intake [Rommers and Meijerhof 1998]. Also, rabbits housed in large groups showed lower feed intake due to a higher level of stress and aggressiveness [Maertens and Van Herck 2000]. In contrast, Matics *et al*. [2018] found no effect of housing system [size of group] on feed intake. The related greater activity in bigger groups affected growth rate and made it slower [Lambertini *et al.* 2001, Dal Bosco *et al.* 2002]. Nevertheless, the greatest body weight and weight gain were observed in the individual housing system [Xiccato *et al*. 1999]. The higher body weight gain was recorded in cage-housed rabbits in contrast with pen-housed rabbits resulting in a better feed conversion ratio in rabbits aged from 7 to 9 weeks [Matics *et al.* 2018]. However, no effect of group size on weight gain or final weight was found [Princz *et al.* 2009, Szendrő *et al*. 2009]. Some authors [Princz *et al.* 2009, Szendrő *et al*. 2009, Combes *et al*. 2010] showed a decline in daily weight gain ranging between 1.0 and 9.3 g/day]. Matics *et al.* [2014] observed no changes in rabbits reared in small groups. On the other hand, Matics *et al*. [2019] found significantly better results of final body weight in rabbits housed in smaller groups.

When stocking density was reduced from 20-23 to 15-16 rabbits/m^2 growth performance would improve [Morisse and Maurice, 1997]. This is consistent with the findings reported by Mousa-Balabel [2009] and El-Bayoumi *et al*. [2018], who observed the lowest body weight gain and body weight in rabbits kept at a stocking density of 28 rabbits/m^2 compared to those reared at a stocking density of 20 or 12 animals/m^2. Princz *et al.* [2008], Szendrő *et al*. [2009], Szendrő and Dalle Zotte [2011] and Paci *et al*. [2013] found either no or only random effect of reducing stocking density to a lower level than 15-17 rabbits/m^2. Nevertheless, higher stocking densities caused lower feed intake in the fattening period [Morisse and Maurice 1997, Trocino *et al*. 2004].

Different types of floor were examined over the years. When comparing three different floor types [wire-mesh vs. plastic-mesh vs. deep litter] a greater body weight was reported in rabbits [aged from 7 to 10 weeks] reared on the plastic-mesh floor than on wire-mesh floor or deep litter. The difference was also found between plastic-mesh and deep litter in 11-week old rabbits. Better results of body weight gain were found in favour of plastic-mesh floor against deep litter [Gerencsér *et al.* 2014]. Trocino *et al.* [2015] observed a higher daily weight gain, feed intake and live weight in the case of plastic floor when compared to wooden slatted floor. When comparing wire-mesh and steel slats floors, greater feed efficiency was found in the case of wire-mesh flooring [Trocino *et al*. 2004]. Indeed, Dalle Zotte *et al*. [2009] compared wire-mesh and plastic-mesh floors with no significant differences in growth performance, which is in accordance with the observations by Dal Bosco *et al.* [2015]. From the productive point of view, deep litter caused a decrease of weight gain and body weight because of its consumption, which has a negative effect on the intake of pellets [Lambertini *et al*. 2001, Matics *et al.* 2014]. Moreover, the effect of using deep litter on a reduction of productive performance was found by Dal Bosco *et al*. [2015].

Carcass traits

The effect of group size on carcass traits was confirmed by several authors [Dal Bosco *et al.* 2002, Dalle Zotte *et al.* 2009, Combes *et al.* 2010, Matics *et al*. 2014]. Lower slaughter and carcass weight, carcass adiposity and greater development of hind parts were reported in rabbits housed in larger groups. This may be caused by increased locomotor activity of rabbits in a bigger space, where the opportunity to move and run is greater [Combes *et al*. 2010]. Also, the dressing out percentage is lower [Dal Bosco *et al.* 2002, Dalle Zotte *et al.* 2009]. Similarly, it also caused a lower fat deposition and a decrease in the meat-to-bone ratio [Combes *et al*. 2010]. A low percentage of dissectible fat in the carcasses was observed in rabbits reared in the outdoor system than in cage-housed rabbits. It is substantiated by a greater energy disbursement involved in moving, jumping and running [Loponte *et al.* 2018]. Some controversial results were published by Machado *et al*. [2019], who observed no effect of housing system [cage vs. pen] and group size [3 vs. 6 rabbits per housing system] for carcass yield, dissectible fat and hind leg yield. They reported the effect of adaptation to the floor system over time. On the other hand, Metzger *et al*. [2003] found significantly better results for carcass yield and slaughter weight in favour of rabbits reared in pens, while Matics *et al*. [2018] reported greater hind parts in pen-raised rabbits.

The stocking density exceeding 15-17 rabbits/m^2 caused an increase in the dressing out percentage [Trocino *et al*. 2004]. At a reduction of the stocking density from 16 to 12 rabbits per cage their carcass weight significantly increased [Trocino *et al*. 2015]. The best parameters of carcass traits were found at a stocking density of 5 rabbits/m^2. The highest skin percentage was found in the housing system of 16 rabbits/m^2 [Paci *et al.* 2013]. There are reports in scientific literature made by Dal Bosco *et al*. [2000] and Pla [2008] showing a trend towards a decreased hind leg proportion when the stocking density increases. That is consistent with statements of Matics *et al*. [2018] on greater higher hind part development, which is favoured by consumers [Dal Bosco et al. 2002].

The effect of floor type on carcass traits was not observed as significant by Princz *et al.* [2009]. On the other hand, Trocino *et al.* [2015] found greater live and carcass weights and dressing out percentage in rabbits reared on the plastic floor compared to those kept on the wooden floor, with these rabbits also having higher muscle-to-bone ratios in hind legs. Dressing out percentage was significantly higher in the case of the wire net floor in comparison with steel slat, plastic slat, wire net and straw litter on wire net floors [Trocino *et al*. 2008]. However, a statement of Dal Bosco *et al.* [2002] also needs to be reported here: "Only when growth is greatly lowered due to unsuitable floors, carcass and meat quality traits are also impaired."

Meat quality – physical and chemical properties

Rabbit meat is a rich source of proteins and essential amino acids and has a high nutritional value. Saturated fatty acids [SFAs] and polyunsaturated fatty acids

[PUFAs] are the most common acids in rabbit meat. The health-promoting value of meat depends on SFAs and fat. The effects of housing systems will be summarised in this section to elucidate the problems. The effect of group size on meat traits such as final pH [24 h *postmortem*] or meat colour was found by several authors [Dal Bosco *et al.* 2002, Dalle Zotte *et al.* 2009, Combes *et al.* 2010, Xiccato *et al.* 2013, Matics *et al.* 2018], who observed the effect of different stress levels on meat colour. Aggressive behaviour and related stress in pen-housed rabbits resulted in the response affecting their muscles, which changed colour due to the lower pH values [Matics *et al.* 2018]. In contrast, Lambertini *et al*. [2001] found no effect of group size on meat colour. The pH values were higher in the *longissimus thoracis et lumborum* muscle in cage-housed rabbits [Dal Bosco *et al*. 2002, Dalle Zotte *et al.* 2009]. However, no changes of pH were found by Combes *et al*. [2010], Xiccato *et al*. [2013] and Palka *et al.* [2018], whereas Lazzaroni *et al.* [2009] observed higher pH in *biceps femoris* and *longissimus lumborum* in pen-housed rabbits due to the capture of rabbits, when they were caught for slaughter. Szendrő and Dalle Zotte [2011] reported the effect of group size on redness [a*] and yellowness [b*] values as unclear. To be exact, Dal Bosco *et al.* [2002] and Dalle Zotte *et al.* [2009] found that *L*a*b** colour values were higher in cage-housed rabbits. In contrast, Combes *et al.* [2010] and Mattioli *et al*. [2016] found these values to be lower in rabbits reared in cages. Szendrő and Dalle Zotte [2011] offered the explanation that the lightness [*L**] value will not change if the pH is not affected by housing system [group size]. Dal Bosco et al. [2002] stated that when the amount of SFAs and monounsaturated fatty acids [MUFAs] in body fat increases, the levels of PUFAs will also increase. Szendrő and Dalle Zotte [2011] explained this trend as the effect of housing of larger groups of rabbits with a decreasing meat lipid content, resulting in an increase in the relative amount of PUFAs.

According to Szendrő and Dalle Zotte [2011], the effect of stocking density on meat composition is not entirely clear, because of examining only stocking densities lower than 17 rabbits/m^2. Following this statement, Preziuso et al. [2009] found higher values for *a** and lower for *L** in rabbits reared at a stocking density of < 5 rabbits per m^2. Matics *et al.* [2014] compared 10.5 rabbits/m^2 and 16.3 rabbits/m^2 finding no differences in colour values of the *longissimus dorsi* muscle. These results are in conflict with those of Dalle Zotte *et al*. [2009] and Paci *et al*. [2013], who found higher *L** values in rabbits housed in higher stocking densities. More researches should be done to examine exact values within a wider range. The effect of stocking density [10 vs. 4 rabbits/m^2] on total PUFA and MUFA contents was reported by Volek *et al*. [2014] in favour of higher stocking density. The floor type could also affect the fatty acid profile in muscles. According to Dal Bosco *et al*. [2015], differences may be observed when comparing wire mesh, plastic mesh and deep litter flooring. Lower amounts of MUFAs in *m. longissimus thoracis* et *lumborum* were detected in the case of the wire mesh floor. Lower PUFA levels were recorded in rabbits kept on the deep litter floor. These values can be discussed only when comparing similar studies. For example Dalle Zotte *et al.* [2009] found that PUFAs did not change in two different

types of housing [cage vs. pen] with no significant effect of floor type [wire mesh vs. plastic net]. In contrast, Dal Bosco *et al.* [2002] recorded a significantly higher level of PUFAs in *longissimus lumborum* and Chodová *et al*. [2014] reported higher PUFA contents in straw bedded rabbits than in caged ones.

Muscle fibres properties

In terms of meat composition, Lefaucher [2010] reported that muscles consist of muscle fibres, type I [βR] and type II [αR, αW]. In rabbits two most dominant muscles are mostly examined [*biceps femoris* and *longissimus thoracis et lumborum*]. Muscle fibres affect the development of postmortem changes, while meat quality is also influenced [Hernández *et al.* 2006]. Two basic characteristics which define muscle fibres are their diameter and perimeter. The size of muscle is also affected by these two characteristics, with cross-sectional area being the third determining factor [Chodová *et al.* 2014]. The effect of stocking density on muscle fibre characteristics in the *biceps femoris* muscle was evaluated by Volek *et al*. [2014]. In rabbits kept at the stocking density of 10 rabbits/m^2 vs. 4 rabbits/m^2 the proportion of αW fibres was 79.3 vs. 59.2%. Comparing the same densities, the proportion of αR fibres was 24.5 vs. 14.2%. Likewise, the distribution of βR fibres was higher at a lower stocking density and amounted to 16.3%, while at a higher stocking density it was only 6.5%. These results were explained by the higher physical activity of the rabbits. The area of βR type muscle fibers was significantly lower in rabbits kept at a lower stocking density [1882 vs. 2744 μm^2], whereas the area of αR and αW fibers was almost unchanged due to stocking density. Very few studies were published on the subject. Chodová *et al.* [2014] discussed two housing systems [collective wire net cages vs. straw-bedded pens] and the resulting development of *biceps femoris* muscle fibres. A different nomenclature of muscle fibre types was used in that study, with type I comprising βR muscle fibres and type II including αR and αW muscle fibres. The trend towards a larger cross-sectional area of muscle type II in comparison with muscle type I was observed in caged rabbits [Gondret *et al*. 2002, Dalle Zotte *et al*. 2005, Chodová *et al.* 2014]. The cross-sectional area of βR fibres [type I] was significantly bigger in rabbits reared in cages at higher stocking densities, with the muscle fibre diameter also being bigger when compared to the cross-sectional area and the diameter of muscle fibres [*biceps femoris*] in rabbits reared at lower stocking densities.

Environmental enrichment of housing systems

Several types of environmental enrichment and their effect on productive or carcass traits were examined in scientific studies, e.g. gnawing sticks [Rizzi *et al*. 2008, Buijs *et al.* 2011, Zucca *et al.* 2012], mirrors [Reddi *et al*. 2011, Musco *et al.* 2019] or platforms [Farkas *et al*. 2016, Matics *et al.* 2018]. The effect of gnawing sticks on productive performance and carcass traits was observed by Hesham and Nasr [2016] indicating better body weight at slaughter, total weight gain, daily feed intake and higher carcass weight. Rizzi *et al*. [2008] found an improvement only in

Table 1. Evaluation of used housing systems with different environmental enrichment types along with their effect on production and welfare of growing rabbits

Authors	Enrichment	Housing system	Effect on production	Effect on welfare
Rizzi *et al*. [2008]	gnawing sticks	individual cages	higher feed intake and growth rate	no data
Buijs *et al*. [2011]	gnawing sticks	open-top wire cages	no data	less social contact, cage manipulation and lateral lying
Zucca *et al*. [2012]	gnawing sticks	enriched cages	low effect on productive performance and meat quality	increased allogrooming
Trocino *et al*. [2013]	gnawing sticks	individual cages x bicellular cages x open-top collective cages	no data	eliminated biting, licking barns or aggressive behavior
Hesham and Nasr [2016]	gnawing sticks	individual cages	better body weight at slaughter, total weight gain, daily feed intake and greater carcass weight	no data
Reddi *et al*. [2011]	mirrors	individual cages	higher body weight gains	higher activity
Mastellone *et al*. [2019]	mirrors	free range	affected energy balancing and consequent productive performance	higher allogrooming, changed behavioral repertoire of isolated rabbits
Musco *et al*. [2019]	mirrors	free range	better growth performance and carcass traits	lower activity, changed behavioral repertoire of isolated rabbits
Farkas *et al*. [2016]	platforms	pens with (plastic or wire-mesh) or without platforms	no significant effect on productive performance due to greater movement in pens in general	no significant effect on welfare
Matics *et al*. [2018]	platforms	pens with (plastic or wire-mesh) or without platforms	no significant effect on productive performance due to greater movement in pens in general	lower frequency of being under platform than in front of them due to urinating by rabbits being on platforms
Trocino *et al*. [2019]	platforms	collective pens	no significant effect on productive performance due to greater movement in pens in general	longer time of resting, being in stretched position and biting or licking objects

feed intake and weight gain. In contrast, Princz *et al*. [2008], Buijs *et al.* [2011] and Zucca *et al*. [2012] found no effect of gnawing sticks on productive and carcass traits. Nevertheless, gnawing sticks should be installed for their benefit of eliminating biting, licking the cage or aggressive behaviour [Trocino *et al*. 2013]. Musco *et al.* [2019] recommended using mirrors in free range rearing rabbits. Placing mirrors in the raising area led to improvement of carcass traits, increased growth performance and dressing out percentage. Rabbits could focus their energy on exploring mirrors, which decreases locomotion activity and increases parameters of live performance traits. In an individual housing system, mirrors had the effect on productive performance, particularly higher growth rate [Reddi *et al*. 2011]. Specifically, mirrors could have an

effect on unwanted behaviour and could eliminate the feeling of isolation [Mastellone *et al.* 2019]. Platforms are generally the most common type of environmental enrichment. While their effect on productive performance is still being investigated, platforms are used to provide environmental enrichment to eliminate agonistic or stereotypical behaviour [Matics *et al*. 2018]. Until recently several studies indicated no significant effects of using multilevel platforms on growth performance [Princz *et al*. 2009, Farkas *et al.* 2016, Matics *et al*. 2018]. When introducing platforms to the cage their correct position inside the cage or pen must be selected to eliminate unhygienic conditions [Trocino *et al*. 2019].

Conclusion

Considering the productive and carcass performance traits, better values of daily weight gain, feed intake, live weight, carcass weight, dressing out percentage and muscle-to-bone ratios were observed in the case of the plastic floor than the wooden slats floor and deep litter. Generally, deep litter caused a reduction of weight gain and body weight, because rabbits consumed the deep litter material. A greater dressing out percentage was found in the case of the wire net floor in comparison with the other floor types. Housing growing rabbits at high stocking densities caused the occurrence of aggressive behaviour, pale meat and lower muscle fibre characteristics. Nevertheless, the best parameters of carcass traits were found at a stocking density of 5 rabbits/m2. Meat quality, such as pH values and lightness, are correlated. If the pH values do not change as a result of the adopted housing system, lightness will be comparable. After the years, scientific literature sources are not consistent in terms of the effect of different housing systems on meat quality in growing rabbits. Very few research studies have evaluated how different rearing systems influence muscle fibre properties. The cross-sectional area, which determines muscle size, is mostly developed in rabbits reared in cages. βR muscle fibres are more developed in housing systems with lower stocking densities, because of greater locomotion activity in these rabbits. The environmental enrichment is very important to reduce stereotypical behaviour. Its effect on productive or carcass traits has not been completely elucidated. Some studies reported no influence of environmental enrichment, whereas different studies informed on greater body weight or better daily feed intake in the case of gnawing sticks and better growth performance in the case of mirrors used for free ranged rabbits. Gnawing sticks should be installed to avoid aggressive behaviour in growing rabbits after sexual maturity. Also mirrors could reduce abnormal behaviour, such as feeling isolated, especially in single-housed rabbits. According to literature research, housing smaller groups of rabbits at stocking densities of 5 rabbits/m2 on the plastic floor with multilevel platforms placed in the middle of the housing system could be recommended.

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How can housing system affect growth and carcass traits, meat quality and muscle fiber characteristics in *biceps femoris* and mineral content of tibia and femur bones in growing rabbits?

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HIGHLIGHTS

- Penned rabbits and males had bigger hind part to reference carcass ratio
- Penned males had the largest αR muscle fiber cross-sectional area and βR diameter
- The higher content of ash and Mg was in the tibia and femur bones of penned rabbits
- Bone fracture toughness was higher in penned rabbits in the tibia and femur bones

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ABSTRACT

The aim of this study was to evaluate the effect of housing system (cages with wire mesh floor and pens with plastic mesh floor) and gender on growth performance, carcass and physical traits, muscle fibers and bone characteristics of growing rabbits. 160 rabbits were assigned to 4 groups by housing system and by gender. Sixty rabbits were kept in cages (0.15 m^2 per rabbit; 90 × 50 × 45 cm; 3 rabbits/replicate; 10 replicates/gender), the other rabbits in pens (0.15 m^2 per rabbit; 25 rabbits/replicate; 2 replicates/gender). Significant housing system (HS) × gender (G) interaction ($P = 0.029$) was found for dressing percentage, when pen housed females showed higher values compared to pen housed males. There was a higher percentage of hind part to reference carcass in pen housed than in cage housed rabbits ($P = 0.001$). The fiber cross-sectional area of αW (white fast twitch) was higher in pen housed ($P = 0.009$) than in cage housed rabbits in *biceps femoris* (BF). Pen housed males had larger diameter ($P = 0.001$) of BF muscle fibers in comparison with pen housed females. Additionally, pen housed rabbits had larger diameter ($P = 0.001$) of BF muscle fibers in comparison with cage housed rabbits. Moreover, significant interactions between HS × G were found in dry matter ($P = 0.010$), calcium ($P = 0.024$) and phosphorus ($P = 0.049$) content in tibias. Pen and cage housed males had higher content of dry matter compared to pen and cage housed females. Regarding calcium results, the highest values were observed in males and females from cages and from penned females, while penned males had the lowest values. The same results were found for phosphorus values. Furthermore, significant HS × G interaction was reported in dry matter content ($P = 0.009$), where the highest values were in pen housed males and the lowest in pen housed females in femurs. Additionally, the higher fracture toughness was found in pen housed rabbits.

1. Introduction

Nowadays, there is a trend to harmonise production with animal welfare by using enriched pen housing systems (Matics et al., 2018). Unfortunately, the human ideas of the ideal housing conditions of rabbits may not correlate with the genuine needs of it (Matics et al., 2014). Generally speaking, consumers want to have an opportunity to buy attractive and implicitly wholesome carcasses produced from welfare-friendly farms (Lazzaroni et al., 2009). Also, the misgivings of an inexpert public are caused by having rabbits as pets (Szendrő et al.,

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2019). Breeders have to be able to satisfy their customers by the high quality of the meat and also demonstrate different housing systems which are more suitable for rabbits than cages (Loponte et al., 2018). There is no generally applicable legislation governing the breeding of rabbits (Szendrő et al., 2019).

In scientific literature, there are no consistent data to clearly state that better results of carcass traits are for cage housed or pen housed rabbits. Changes in carcass yield could be related to higher movement in pens (Matics et al., 2019). Metzger et al. (2003) found significantly better results for carcass yield and slaughter weight in favor of cage housed rabbits and Matics et al., (2018) stated a higher level of hind parts in pen housed rabbits, but the reference carcass yield was higher in caged rabbits.

Muscle fiber characteristics influence the physical and sensory properties of meat (Dalle Zotte et al., 2005). The presence of muscle fibers affects the course of *postmortem* changes and also the quality of meat (Hernández et al., 2006). Meat quality depends also on structure and composition of muscle fibers (Blasco and Ouhayoun, 1996). Muscle fibers are described according to the methodology of Ashmore and Doerr (1971), who divided muscle fibers according to contractile and metabolic characteristics into types αR (red, rapidly contractile), αW (white, rapidly contractile) and βR (red, slowly contractile). The number of muscle fibers per 1 mm^2 is closely related to the area of the fibers (Ouhayoun and Dalle Zotte, 1993). The transformation of muscle fibers is reversible, in the case of higher muscle work, the muscle fibers may change from white muscle fibres back to αR, as the number of mitochondria increases in the fibers. Rabbits with a higher possibility of locomotor activities show a higher percentage of βR and αR fibers and a decrease in the proportion of αW fibers (Volek et al., 2014). Higher locomotion is also connected with higher fracture toughness of bones (Dalle Zotte et al., 2009) and mineral content (Sharma et al., 2021), which is as important as content of ash and dry matter in occurrence of osteoporosis in adult animals.

The aim of this study was to evaluate the effect of housing system (cages with wire mesh floor and pens with plastic mesh floor) and gender and their interactions on growth performance, carcass and histomorphological traits, muscle fibers and bone characteristics of growing rabbits.

2. Materials and methods

The study was realized in the Demonstration and Experimental Centre of the Faculty of Agrobiology, Food and Natural Resources, Czech University of Life Sciences Prague (Czech Republic). The experiment was authorized by the Ethical Committee for Animal Experimentation of Czech University of Life Sciences Prague. The observation was in agreement with the attitude stated in Directive 2010/63/EU revising Directive 86/609/EEC on the protection of animals used for scientific purposes.

2.1. Animals and experimental design

The rabbits were housed in a room with controlled environmental conditions. The ventilation and rating system allowed an ambient temperature to be maintained between 18 and 21°C, and relative humidity between 65-70% throughout the experiment period. The daily lighting period was 12 h. The rabbits were fed ad libitum by a feeder with a standard fattening pelleted diet containing 14.5% crude protein and 10.2 MJ of digestible energy/kg. Water was available ad libitum from nipple drinkers.

A total of 160 rabbits of commercial hybrid Hyplus (PS 19 × PS 39) of both genders (1:1) were weaned at 36 d of age. They were randomly divided into 4 experimental groups (by housing system and by gender). Sixty rabbits were kept in cages with wire mesh floor made from galvanized wire with 3 mm diameter (0.15 m^2 per rabbit; 90 × 50 × 45 cm; 3 rabbits/replicate; 10 replicates/gender), the other rabbits were kept in pens with plastic mesh floor with 10 mm slat width, 10 mm slot width, and 50% perforation and the walls were wire meshed (0.15 m^2 per rabbit; 25 rabbits/replicate; 2 replicates/gender). There was an elevated step in the middle of the pen, which divided the pen to two halves. The step was elevated above the second half by 15 cm.

2.2. Growth performance, carcass and physical characteristics, sampling and analysis

Individual live weight was recorded weekly and feed intake was noted once per day in both housing systems to calculate body weight gain, feed intake and the feed conversion ratio.

All rabbits were weighed at the end of the (80 d of age). 15 rabbits per experimental group were randomly selected for carcass and other analysis. Selected rabbits were then weighed (SW) and slaughtered.

Slaughter and carcass dissection progresses were evaluated in accordance with the norms of the World Rabbit Science Association recommendation by Blasco and Ouhayoun (1996). The slaughtered rabbits were bled, and then the skin, genitals, bladder, gastrointestinal tract, and the distal segment of the legs were removed. Twenty minutes after slaughter, carcasses with the head, liver, kidneys, perirenal and scapular fat and set of organs consisting of thymus, trachea, esophagus, lungs and heart were weighed (hot carcass; HC). They were suspended from the tendon calcaneus for 1 h in a ventilated area and then chilled at 4°C for 24 h. After 24 h, the carcasses were weighed to obtain the chilled carcasses (CC). The cooler carcass shrinkage (CCS) was calculated as the difference between the hot carcass and chilled carcass, relative to the hot carcass. The head, liver, kidneys and set of organs, were removed from each carcass to obtain the reference carcass (RC), which included the meat, bones, and fat deposits. The carcasses were then cut between the 7^{th} and 8^{th} *thoracis vertebrae* and between 6^{th} and 7^{th} *lumbar vertebrae* to obtain the fore, mid, and hind parts, which were weighed separately. Next, the hind part was obtained by the technological division (Blasco and Ouhayoun, 1996) and the bone and meat of the hind legs were dissected, and their weights were recorded. The dressing percentage (DP; CC divided by SW and multiplied by 100) and the ratio of the organs and carcass parts to either the CC or to the RC were calculated as required. The head, lung, liver and kidneys weights were expressed as a percentage of the CC. The fore, mid and hind parts, thigh, thigh muscle and dissectible fat weights were expressed as a percentage of the RC.

The ultimate pH (pHu) of the *biceps femoris* (BF) was measured 24 h *postmortem* using a pH meter WTW pH 330i (WTW, Weilheim, Germany) provided with a glass electrode suitable for meat penetration.

Muscle color regarding, the aperture size was 8 mm and it was the diameter of measured area, a specular component 0% UV, standard illuminant D65 (daylight simulation), observer angle was 10°, and zero and white calibration values corresponded to the average of three measurements per sample. The L* a* b* color values were measured 24 h *postmortem* on the fresh cut surface. Meat color of the hind leg was measured in transversal section of the BF muscle surface. Instrumental color measurements were recorded in the CIELAB color space for L* (lightness – 0 = black, 100 = white), a* (redness/greenness - positive values = red, negative values = green), and b* (yellowness/blueness - positive values = yellow, negative values = blue) using spectrophotometer (CM-700d, Konica Minolta, Osaka, Japan). The measurements were taken after 30 min of air exposure to allow blooming.

Then, the hind leg meat was dissected and collected to determine the BF muscle fiber characteristics. Samples of BF muscle were frozen in liquid nitrogen-cooled isopentane (-156°C) and then stored at -80°C until analysis. Serial cross-sections (12 μm) from BF were obtained with a cryostat Leica CM1850 (Leica Microsystems Nussloch GmbH, Nussloch, Germany) at -20°C. The sections were subjected to myofibrillar ATPase staining after successive preincubations in acid (pH = 4.6) and alkaline buffer (pH = 10.3), according to methodology by Brooke and Kaiser (1970). The computerized images analysis of fibers were typed as βR (red slow twitch fiber), αR (red fast twitch fiber) or αW (white fast

twitch fiber) according to the nomenclature described by Ashmore and Doerr (1971). For each muscle fiber type, the number in 1 mm^2, fiber type distribution (%), diameter (μm), perimeter (μm), cross-sectional area (μm^2) and circularity were determined, using NIS Elements AR 3.1 software (supplied by Laboratory Imaging s.r.o., Prague, Czech Republic).

2.3. Bone characteristics, sampling and analysis

The bones of the hind leg (femur and tibia) were individually packed in polyethylene bags and stored at -20°C until analysis, when they were thawed overnight. When fully thawed, soft tissue was removed from the tibia and femur. The tibia and femur were subsequently boiled for 15 min in 95°C water, defleshed further and dried at 25°C for 24 h. Subsequently, the maximum shear force until initial structural silure (i.e. the breaking of the bone) was determined to a three-point flexure test using a Instron® Model 3342 (Instron, Norwood, Massachusetts, US) and the load rate was 12 mm/min. The distance between the two fulcra points supporting the bones was 45 and 38 mm. The bones were constantly oriented for testing with their natural convex shape downwards. The bone dry matter content was determined by oven drying at 105°C. The ash content was determined by oven burning at 550°C. After burning the sample in the oven, the sample was decomposed (7 ml HNO_3 + 1 ml HF supplemented with water to a volume of 50 ml). Bone calcium (Ca), phosphorus (P) and magnesium (Mg) were determined in ash samples: Ca and Mg by atomic absorption spectrometry on device ContrAA 700 and P colorimetrically by a molybdate reagent (Huxtable and Bressler, 1973).

2.4. Statistical analysis

The statistical analysis was processed using the software SAS 9.4, 2012 (SAS Institute Inc. Cary. NC. USA). All the data were examined using the General Linear Model with PROC MIXED procedure:

$$Y_{ijk} = \mu + HS_i + G_j + (HS \times G)_{ij} + e_{ijk}, \text{where}$$

Y_{ijk} value of trait,
μ general mean,
HS_i effect of housing system (i = 1-2),
G_j effect of gender (j = 1-2),
$(HS \times G)_{ij}$ effect of interaction of level i of housing system with level j of gender, e_{ijk} random residual error.

Housing system and gender were considered as fixed effects. Differences between means were determined by the Duncan's test. The value of $P \leq 0.05$ was considered significant for all measurements. All the data are expressed in tables as LS means. The interaction effect and mean values were not reported in tables but discussed in the text when significant.

3. Results

3.1. Growth performance characteristics

Results of growth performance are presented in Table 2. Housing system significantly affected live weight at 80 d, body weight gain and feed intake. These characteristics were observed higher in cages. Moreover, effect of gender showed significant differences between males and females in live weight at 80 d and in body weight gain, when both parameters were higher in females.

3.2. Carcass characteristics

Some carcass characteristics were more affected by the housing system, than by the gender (Table 3). The cage housed rabbits (housing system effect; HS) and females (gender effect; G) showed a higher slaughter weight (+211 g and +175 g, resp.) compared to other rabbits. The full digestive tract was influenced by the housing system, gender and there was found significant HS × G interaction. Cage housed females showed a higher full digestive tract by 144 g, 140 g and 100 g in comparison with pen housed males or females and cage housed males. Significantly higher values of full digestive tract were found in cage housed rabbits in comparison with pen housed rabbits. Similarly, significant HS × G interaction was stated for dressing percentage, when pen housed females showed the highest dressing percentage (61.1%) compared to other ones. None of the studied factors or HS × G interaction had a significant influence on hot carcass, chilled carcass, reference carcass and cooler carcass shrinkage, as well as most of the selected ratios of organs and carcass parts to reference carcass. Significantly heavier head and lung to chilled carcass ratio were observed in pen housed rabbits compared to cage housed rabbits.

There was a higher hind part to reference carcass ratios in pen housed rabbits (+1.6 percentage points) than in cage housed rabbits. Likewise, the higher hind part to reference carcass ratio was higher in females (by +1.0 percentage points) than in males. Similarly, the pen housed rabbits demonstrated the both significantly higher thigh to reference carcass ratio (+0.7 percentage points) and thigh muscle to reference carcass ratio (+0.5 percentage points) in contrast to cage housed rabbits.

Table 1
Ingredient and chemical composition of pelleted diet (%).

Item Ingredient	%
Alfalfa meal	30
Sunflower meal	17
Wheat bran	23.5
Sugar beet pulp	4
Oats	13
Barley	8
Rapeseed oil	2
Vitamin-mineral premix[a]	0.5
Monocalcium phosphate	0.5
Limestone	1
Salt	0.5
Chemical composition	
Dry matter	88.6
Crude protein	14.5
NDF[c]	29.4
ADF[c]	18.3
ADL[c]	3.9
Ether extract	3.0
Starch	18.7
Ash	6.6
Ca	0.996
P	0.649
Mg	0.308
Digestible energy (MJ/kg)[b]	10.2

[a] Included per kg of diet: vitamin A, 12,000 IU; vitamin D3, 1500 IU; vitamin E, 50 mg; vitamin K3, 1 mg; vitamin B1, 2 mg; vitamin B2, 6 mg; vitamin B6, 3 mg; niacinamide, 20 mg; Ca-pantothenate, 10 mg; folic acid, 0.5 mg; vitamin B12, 0.02 mg; choline chloride, 500 mg; Co, 0.3 mg; Cu, 8 mg; Fe, 27 mg; I, 0.8 mg; Mn, 19 mg; Zn, 44 mg; Se, 0.07 mg.
[b] As provided by the compound feed manufacturer.
[c] NDF: neutral detergent fibre, ADF: acido detergent fibre, ADL: acido detergent lignin.

3.3. Physical measurements of biceps femoris muscle

Table 4 presents pHu and color parameters (L* a* b*) of BF muscle. Housing system significantly affected only b* color values of the BF muscle (by +1.11 in favor of pen housed rabbits).

Table 2
Growth performance according to housing system and gender.

Traits	Housing system (HS)		Gender (G)		SEM[a]	*P*-value[b]		
	Pen	Cage	Male	Female		HS	G	HS × G
Live weight 36 d (g)	919	896	900	915	11	0.302	0.520	0.330
Live weight 80 d (g)	2793	3017	2848	2984	34	0.005	0.039	0.210
Period 36 – 80 d								
Body weight gain (g/d)	42.6	48.3	44.3	47.1	0.7	0.001	0.020	0.219
Feed intake (g/d)	140	148	147	147	5.94	0.043	0.089	0.662
Feed conversion ratio	3.30	3.15	3.18	3.11	0.09	0.652	0.673	0.375

[a] SEM = Standard error of the mean.
[b] *P*-values were considered significant at 0.05 level.

Table 3
Effect of housing system and gender on selected slaughter traits.

Traits[c]	Housing system (HS)		Gender (G)		SEM[a]	*P*-value[b]		
	Pen	Cage	Male	Female		HS	G	HS × G
Slaughter weight (SW, g)	2759	2970	2777	2952	46	0.017	0.045	0.697
Skin (g)	403	436	412	427	8	0.039	0.330	0.194
Full digestive tract (g)	407	498	426	479	14	0.001	0.016	0.026
Hot carcass (HC, g)	1700	1817	1709	1808	30	0.052	0.097	0.784
Chilled carcass (CC, g)	1674	1790	1683	1780	30	0.051	0.101	0.832
Reference carcass (RC, g)	1377	1474	1378	1473	27	0.063	0.069	0.716
Cooler carcass shrinkage (CCS, %)	1.50	1.50	1.50	1.50	0.05	0.581	0.896	0.091
Dressing percentage (DP, %)	60.6	60.3	60.6	60.3	0.28	0.510	0.647	0.029
Head (% CC)	7.80	7.20	7.80	7.30	0.13	0.029	0.051	0.908
Lung (% CC)	0.90	0.70	0.80	0.80	0.03	0.038	0.624	0.755
Liver (% CC)	6.00	6.51	6.40	6.12	0.20	0.111	0.314	0.984
Kidneys (% CC)	1.21	1.10	1.14	1.07	0.02	0.084	0.144	0.990
Fore part (% RC)	42.2	42.2	42.4	42.1	0.26	0.949	0.618	0.554
Mid part (% RC)	18.4	18.2	18.0	18.5	0.17	0.596	0.205	0.949
Hind part (% RC)	36.6	35.0	36.3	35.3	0.25	0.001	0.036	0.737
Thigh (% RC)	17.4	16.7	17.3	16.9	0.12	0.001	0.098	0.807
Thigh muscle (% RC)	13.4	12.9	13.3	13.0	0.09	0.002	0.071	0.701
Kidney fat (% RC)	1.85	2.22	1.89	2.17	0.11	0.101	0.216	0.995
Inguinal fat (% RC)	0.61	0.61	0.49	0.74	0.07	0.988	0.060	0.488
Scapular fat (% RC)	0.58	0.65	0.58	0.65	0.05	0.460	0.459	0.609
Total dissectible fat (% RC)	3.04	3.48	2.96	3.56	0.16	0.162	0.060	0.900

[a] SEM = Standard error of the mean.
[b] *P*-values were considered significant at 0.05 level.
[c] HC = carcass after slaughter with head, set of organs consisting of thymus, trachea, esophagus, lungs and hearth, liver, kidneys, perirenal and scapular fat; CC = chilled carcass weight 24 h after slaughter; RC = CC minus the head, liver and kidneys, thoracic cage organs and neck (RC included the meat, bones, and fat deposits); CCS = difference between HC and CC divided by HC (× 100); DP = CC weight divided by slaughter weight and multiplied by 100; Total dissectible fat includes the scapular, inguinal and perirenal fat.

Table 4
Effect of housing system and gender on some physical characteristics of *biceps femoris* muscle.

Traits[c]	Housing system (HS)		Gender (G)		SEM[a]	*P*-value[b]		
	Pen	Cage	Male	Female		HS	G	HS × G
pHu	5.76	5.71	5.75	5.71	0.01	0.096	0.147	0.137
L* value	61.9	60.8	61.6	61.1	0.57	0.320	0.710	0.111
a* value	-2.58	-3.07	-2.67	-2.97	0.14	0.083	0.286	0.330
b* value	7.76	6.65	7.46	6.95	0.26	0.034	0.322	0.805

[a] SEM = Standard error of the mean.
[b] *P*-values were considered significant at 0.05 level.
[c] pHu = pH ultimate, 24 h after slaughter; L* = lightness; a* = redness; b* = yellowness.

3.4. Fibre histomorphological characteristics of biceps femoris muscle

The results about the fiber histomorphological characteristics of BF muscle are presented in Table 5. Fiber type distribution was only affected by gender, specifically only the distribution of αW muscle fibers, when females had higher values by +17.3 percentage points than males. Significant differences were found in HS × G interaction for diameter of αR muscle fibres. The pen housed males had the largest diameter (60 μm), while pen housed females had the smallest diameter (55.7 μm). Significant differences were found in HS × G interaction for perimeter as well. The pen housed males had also the largest perimeter of αR muscle fibers (224 μm) compared to pen housed females (203 μm), who had the lowest value of this parameter. The same trends as in αR muscle fibers, the largest perimeter and diameter in the same housing system and the gender, were observed in αW muscle fibres. Cage housed females had the largest perimeter and diameter of αW muscle fibers compared to males in the same housing system (243 μm vs. 226 μm; 65 μm vs. 60 μm, resp.), who had the lowest values. Significant differences were found in HS × G interaction for αR and αW fibre cross-sectional area. Pen housed males had the largest αR fiber cross-sectional area

Table 5
Effect of housing system and gender on the muscle fibre type distribution and fiber histomorphological characteristics of biceps femoris muscle.

Characteristic[c]	Housing system (HS)		Gender (G)		SEM[a]	*P*-value[b]		
	Pen	Cage	Male	Female		HS	G	HS × G
Number of fiber (in 1 mm^2)								
βR	24.4	26.2	27.0	23.6	2.36	0.712	0.487	0.437
αR	130	110	142	97.6	11.68	0.393	0.056	0.374
αW	123	133	108	149	12.92	0.688	0.116	0.142
Total	277	170	277	270	8.97	0.683	0.683	0.466
Fiber type distribution (%)								
βR	8.89	9.45	9.57	8.78	0.83	0.745	0.641	0.151
αR	48.0	41.6	53.1	36.6	4.26	0.444	0.053	0.343
αW	43.1	48.9	37.4	54.7	4.36	0.489	0.046	0.223
Diameter (μm)								
βR	53.6	51.3	52.9	52.1	0.78	0.150	0.635	0.064
αR	57.8	57.1	57.8	57.1	0.44	0.447	0.446	0.001
αW	61.2	62.4	60.6	63.0	0.43	0.159	0.006	0.003
Perimeter (μm)								
βR	193	183	192	184	2.86	0.068	0.019	0.135
αR	213	210	215	208	1.77	0.389	0.049	0.001
αW	232	234	229	237	1.80	0.450	0.020	0.016
Fiber cross-sectional area (μm^2)								
βR	2408	2165	2298	2276	67.73	0.076	0.868	0.109
αR	2807	2743	2825	2724	40.62	0.442	0.221	0.001
αW	3111	3253	3046	3318	40.70	0.088	0.001	0.001
Circularity								
βR	0.77	0.78	0.76	0.79	0.006	0.093	0.002	0.067
αR	0.74	0.74	0.72	0.75	0.002	0.510	0.001	0.260
αW	0.71	0.71	0.71	0.72	0.003	0.357	0.058	0.056

[a] SEM = Standard error of the mean.
[b] *P*-values were considered significant at 0.05 level.
[c] βR = Red slow twitch fiber; αR = Red fast twitch fiber; αW = White fast twitch fiber.

and pen housed females had the lowest values for this parameter (3022 μm^2 vs. 2591 μm^2). Next, cage housed females had the highest of the αW fibre cross-sectional area (3527 μm^2), on the other hand, in cage housed males there were found the lowest values (2979 μm^2). Likewise, the αW fiber cross-sectional area was larger in females by +273 μm^2 in comparison with males. In the present study, the circularity was influenced by the gender, where females had higher circularity of βR and αR muscle fibers than males.

3.5. Tibia and femur bone characteristics

Table 6 provides results of the mineral content and fracture toughness in the tibia and femur.

Regarding tibia bones, housing system significantly affected ash content in favor of pens by + 4.09 %, calcium in favor of cages by +10 g/kg, magnesium with better results in pens (6.94 vs. 6.62 g/kg) and in WBFT, where rabbits from pens had higher fracture toughness by + 43 N.

The gender significantly affected content of dry matter, when males had higher values than females and magnesium, which was higher in females compared to males. Moreover, significant interactions between HS × G were in dry matter content, calcium and phosphorus. Interactions tended to be significant in magnesium. Pen and cage housed males had the highest content of dry matter compared to the lowest dry matter content in cage housed females. Regarding calcium results, the highest values were observed in males and females from cages and from penned females, while penned males had the lowest values. The same trends were found for phosphorus values.

Considering femur bone, housing system affected ash and magnesium content in favor of penned rabbits. Additionally, WBFT tended to

Table 6
Effect of housing system and gender on the mineral content and fracture toughness in tibia and femur bones.

Traits[c]	Housing system (HS)		Gender (G)		SEM[a]	*P*-value[b]		
	Pen	Cage	Male	Female		HS	G	HS × G
Tibia								
Dry matter (%)	96.6	96.6	97.2	95.9	0.2	0.950	0.001	0.010
Ash (%)	63.6	59.5	60.9	62.7	0.9	0.031	0.413	0.664
Calcium (g/kg)	287	297	289	293	1.7	0.001	0.181	0.024
Phosphorus (g/kg)	179	181	178	181	1.1	0.358	0.198	0.049
Magnesium (g/kg)	6.94	6.62	6.65	6.95	0.1	0.011	0.023	0.057
WBFT (N)	411	368	380	400	8.60	0.007	0.211	0.079
Femur								
Dry matter (%)	96.3	96.5	97.1	95.8	0.2	0.673	0.001	0.009
Ash (%)	60.8	57.6	59.0	59.4	0.4	0.001	0.524	0.499
Calcium (g/kg)	298	298	305	292	2.9	0.993	0.033	0.924
Phosphorus (g/kg)	184	183	187	180	1.7	0.714	0.057	0.503
Magnesium (g/kg)	7.20	6.75	6.87	7.07	0.1	0.003	0.170	0.461
WBFT (N)	307	265	291	281	10.70	0.052	0.654	0.980

[a] SEM = Standard error of the mean.
[b] *P*-values were considered significant at 0.05 level.
[c] WBFT = Warner Bratzler Fracture Toughness.

be significant with results of higher resistance to fracture in pens by + 42 N. Furthermore, males had more bone dry matter content than females. Oppositely, females had higher values of magnesium than males. Phosphorus tended to be higher in males. Likewise, significant interaction between HS × G was in dry matter content, where the highest values were in pen housed males and the lowest in pen housed females.

4. Discussion

4.1. Growth performance characteristics

Generally, the growth performance of rabbits reared in pens is lower compared with cage housing systems with small groups of rabbits (Lambertini et al., 2001; Princz et al., 2009; Xiccato et al., 2013a). A pen provides more free space for locomotion and lower feed intake with lower weight gain (Maertens and Van Herck 2000). This trend was observed also in our study, where higher final weight, body weight gain and feed intake were higher in cages than in pens. In pens, expression of different behavior than in cages can occur. Competitions for feed (Xiccato et al., 2013b), increased movement (Dal Bosco et al., 2002) and energy requirements (Loponte et al., 2018) are the main consequences of pen or free range rabbits. Regarding the gender effect, in the present study was found, that females had higher final live weight and body weight gain. Additionally, Bozicovich et al. (2016) summarized, the lower feed intake and growth performance of males could be affected in the same reason of specific behavior, which can be manifest as aggressive among males.

4.2. Carcass characteristics

In the present study, significantly higher slaughter weights of rabbits in cages and in females were found with notable differences. + 211 g as the difference between rabbits from cages and pens and +200 g between females and males could be explained by higher physical activity of penned rabbits and males. Males' higher activity could be explained by their earlier achievement of sexual maturity and connected aggression (Di Meo et al., 2016). Lazzaroni et al. (2009) found higher differences between cage or pen housed rabbits (+290 g in cages) compared to our results with the explanation of higher locomotor activities of rabbits in pens. Also, they reported higher slaughter weight of females by +101 g. Additionally, Bozicovich et al. (2016) found the lowest number of injured rabbits in female groups, which could indicate more calm among females and due to that lower movement. Regarding the full digestive tract, the highest values were found in females, which correspond with our previously mentioned hypothesis that males' were more active. In present study, females from pens had the highest dressing percentage compared to males from pens and cage housed rabbits. These differences cannot be clearly assigned to the treatment. According to scientific studies (Dal Bosco et al., 2002; Xiccato et al., 2013a), the better performance of rabbits from cages compared to rabbits from pens (collective housing) is related to expression of behavior, ability to movement and affected feed intake. These results could show trends, when rabbits housed in pens with high floor space, they demonstrate heavier lungs, maybe partly caused by higher locomotion and higher air exchange in the lungs. Also, heavier heads in rabbits with higher locomotion activity might be caused due to higher relative brain weight. This tendency was observed by Bozicovich et al. (2016), who found numerically heavier brains in male rabbits housed in enriched systems and confirmed this information with facts that mice and rats housed in enriched systems showed the same trends. All of that could be caused by environmental stimulus and higher energy requirements of animals (Allman, 2000). Considering the higher locomotor activities, it is likely to affect the development of hind part to reference carcass ratio in pen rabbits (Gondret et al., 2009; Lazzaroni et al., 2009; Combes et al., 2010; Matics et al., 2018). This trend is confirmed in our results, where both a higher hind part and thigh to reference carcass ratio were found in pens than in cages.

4.3. Physical measurements of biceps femoris muscle

According to Szendrő and Dalle Zotte (2011), pHu values were measured as lower, when group size of rabbits was bigger. Specifically, agonistic behavior in these groups after reaching sexual maturity can occur and have an impact on rheological traits. Trocino et al. (2019) reported aggression at 9 – 10 weeks of a rabbit's age and Trocino et al. (2014) found higher levels of corticosterone levels in pen housed rabbits. That could indicate lower pHu values due to adaptive stress response in muscles. On the other hand, increased locomotor activities have an effect on αW muscle fibers and transform them to αR fibers, which induce increased pHu values (Ouhayoun, 1998). In our study, higher pHu values of BF in pen housed rabbits and males could be attributed to higher movement of penned rabbits and males, who could expressed their attention against the same gender and induce then the higher locomotion. However, the results did not show the possible aggression among pen housed rabbits and males in pHu point of view, the stressful conditions created by agonistic behavior could induce adaptive response in the muscles for metabolic production of more free radicals (Dal Bosco et al., 2002). According to Dalle Zotte et al. (2009), the pen housed rabbits provided better response to pre-slaughter treatments. In the present study, cage housed rabbits could negatively react to pre-slaughter handling, decreased their pHu and due to that pen housed rabbits had higher values with connection of higher movement as well as males compared to females.

Regarding the meat color, Dalle Zotte et al. (2009) reported the lower the pHu is, the lighter the meat color is. Our results showed numerically higher L* values in penned rabbits and in males in BF muscles. Moreover, b* color values were higher in BF in pens. Contrary to us, Dal Bosco et al. (2002) and Dalle Zotte et al. (2009) found higher values of L* a * b* in cages. On the other hand, Combes et al. (2010) observed same results as we did. However, according to Szendrő and Dalle Zotte (2011) the effect of group size on color values is unclear, Zhao et al. (2018) found positive correlations between b* with pHu values *postmortem*.

4.4. Fibre histomorphological characteristics of biceps femoris muscle

As it was concluded in the review of Krunt et al. (2020), two the most examined muscles on rabbits carcass are *biceps femoris* and *longissimus thoracis et lumborum*, which consists of typically investigated muscle fibers (βR, αR, αW). The diameter and perimeter define muscle fiber and the size of muscle is expressed by cross-sectional area. Ouhayoun (1998) reported transformation of αW to αR muscle fibers due to higher movement. The changes can also occur with the age (Chodová et al., 2018). In the present study, females had more αW muscle fibers, which show males were more active, as was previously mentioned. This trend is displayed in results of αR muscle fiber distribution, which tended to be higher in males. The locomotor activities, specifically in pen males, affected distribution of αR fibers and their diameter and perimeter. Therefore, they had the largest αR fiber cross-sectional area. Oppositely, females in cages had the largest diameter, perimeter and cross sectional area of αW fibers. Choi et al. (2009) correlated increased cross-sectional area of muscle fibers with live weight. Additionally, Lefaucheur (2011) observed, selection of rabbits for growth resulted in increased fibers size. According to available surface area, Volek et al. (2014) compared 10 rabbits/m^2 and 4 rabbits/m^2 with results of αW distribution (79.3 vs 59.2%), which can be attributed to our results in cages, specifically in caged females. The higher cross sectional area in αW and αR was observed in studies of Dalle Zotte et al. (2005) and Chodová et al. (2014), in favor of cage housed rabbits compared to pen housed rabbits. In our study, we observed the same trend in pen males.

4.5. Tibia and femur bone characteristics

Mineralization of bones can be described by calcium, magnesium, phosphorus, and iron content (Clarkson and Haymes, 1995). Calcium and phosphorus are the main elements of the mineral matrix and they vary independently. Most of the calcium content is tied up in the mineral phase and goes to the blood through bone resorption (Peacock, 2010). Moreover, sustaining homeostasis and bone metabolism depends on the relative content of calcium and phosphorus, which is needed for growth and development of bones (Shapiro and Heaney, 2003). Low bone mass may increase risk of fracture. In humans, one of the factors affecting bone mass and its strength is lifestyle with respect to exercise (Clarkson and Haymes, 1995). Running on the treadmill is one form of exercise for rats. Specifically, femur and tibia bones receive a great mechanical loading than the lumbar spine (Iwamoto et al., 2005). Furthermore, exercise promotes a positive calcium balance and increases skeletal mass (Yeh et al., 1989). In laying hens, higher ash percentage contents were found in floor pens (Silversides et al., 2012) and in free range housing (Sharma et al., 2021) compared to cage housing systems. Moreover, higher breaking strength of bones in penned rabbits could be explained by connection with higher ash percentage, which shows mineral contents, such as calcium (Sharma et al., 2021) or magnesium. According to our results, it seems to be magnesium that affects bone strength more than calcium or phosphorus. However, calcium influences bone strength (Zhu and Prince, 2012), magnesium is its antagonist (Iseri and French, 1984) and also has the effect on bone fractures, which was reported in the study of Boskey et al. (1992). Considering dry matter content, males could have higher values due to their higher hind part, which was affected by locomotion and due to that the bones from pens are usually longer and heavier than bones from cages (Matics et al., 2018) and have more dry matter. The differences of single elements between genders could be explained by different locomotor activity, which correspond with behavior, sexual maturity (Di Meo et al., 2016) and connected movement (Combes et al., 2010), as was found in studies of human bones, where reduced exercise decreased mineral content of bones (Vico et al., 2000).

5. Conclusion

The best results of growth performance and carcass can be obtained in cages. In case of distributing parts of carcasses to supermarkets, the highest hind parts were in rabbits housed in pens, which could be valuable. Moreover, higher fracture toughness was observed also in pen housed rabbits. That could indicate higher welfare status in terms of bone fractures. The results showed critical values of minerals in bones of variously housed rabbits and could set the future perspectives of modelling a suitable housing system for rearing young females for future breeding with respect to their bone strength and its welfare to avoid injuries and osteoporosis in adult females.

Declaration of competing interest

We agree that this article is original, is not being considered for publication elsewhere, and it is approved by all authors. There is no conflict of interest.

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The effect of housing system on rabbit growth performance, carcass traits, and meat quality characteristics of different muscles

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ABSTRACT

The present study analysed the effect of housing system (caged versus penned) on the growth performance, carcass traits, and meat quality of rabbits. The physiochemical quality of five muscles was evaluated, together with the chemical composition and fatty acid profile of the meat from the hind leg. Sensory properties of the *longissimus lumborum* (LL) was also assessed. The LL yields were higher in caged rabbits. According to the results, meat from rabbits raised in cages had higher b* values for the LL (i.e., meat was yellower) than rabbits raised in pens. In addition, the *quadriceps femoris* from penned rabbits had higher a* and b* values (i.e., were redder and yellower) than caged rabbits. The MUFAs (18:1 n-9 and 20:1 n-9) and ash contents of the hind leg meat were higher in caged rabbits. In conclusion, while housing system influenced the physiochemical traits of rabbit meat, the sensory properties were not influenced.

1. Introduction

The production and consumption of rabbit meat are rising in countries such as China and Mexico; whilst European countries with a tradition of rabbit production and consumption, such as Italy, Poland, France, and Spain, have noted a significant reduction (FAOSTAT, 2020). Welfare, primarily associated with animal housing conditions, may be the key player regarding the popularity of rabbit meat, particularly in European countries (Petracci, Soglia, & Leroy, 2018). Moreover, recent husbandry changes made in connection with welfare standards (e.g., utilization of group housing for growing rabbits in pens with different construction dimensions and materials) are associated with higher expenses, making rabbit rearing more costly for farmers (Szendrő, Szabó-Szentgróti, & Szigeti, 2020). There are also predictions that cage farming will be completely banned in some countries (e.g., the Czech Republic) in the near future (Valkova et al., 2021). In order to adhere to new welfare guidelines, adapting the entire rabbit farming production cycle to alternative systems will be necessary. Research has shown that housing systems significantly impacts production efficiency and meat quality in rabbits (Combes, Postollec, Cauquil, & Gidenne, 2010; Dalle Zotte et al., 2015; Krunt, Zita, Kraus, & Volek, 2021; Xiccato, Trocino, Majolini, Tazzoli, & Zuffellato, 2013). Recent changes in the housing requirements of rabbits present their own welfare issues, potentially introducing new stressors of social and behavioural origin, and thus research should not neglect to consider the influence of this on the production performance and meat quality of these rabbits.

Several scientific studies have compared the effects of housing rabbits in traditional systems (cages) and alternative systems (pens) on their productive parameters and meat quality. Pla, Zomeño, and Hernández (2008), reported lower growth rates, meat pH, and meat surface redness from pen-housed rabbits compared with cage-housed rabbits. Mattioli et al. (2016) examined the differences between bicellular cages and mobile arks with shelters, and reported no effects on pH, colour, water holding capacity, or cooking loss. On the other hand, the authors found a higher thrombogenicity index and a lower peroxidability index in the rabbit meat from animals in cages. Housing system also affects the chemical composition of rabbit meat, likely due to differences in the level of physical activity possible within each system, influencing muscle development and fat deposition, both of which are important for physiochemical and sensory meat quality. Dalle Zotte et al. (2015)

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reported significantly higher ash contents in meat from rabbits in pens compared to those raised in cages. Szabó, Romvári, and Fébel (2002) showed that differences in meat fatty acids profiles from rabbits under different housing systems are influenced by the degree of movement of the rabbits, with decreased contents of C18:0 and C20:4 n-6 in the hind leg muscle (*vastus lateralis*) of rabbits exhibiting more movement/exercise. Most of these previous studies focused on quality investigations of the two largest muscles in rabbits from the loin and hind leg, i.e., the *longissimus lumborum et thoracis* (LTL) and *biceps femoris* (BF) muscles. However, the *semitendinosus* (ST) or *quadriceps femoris* (QF) muscles are usually not considered but are important muscles in the rabbit's movement. It is important to expand the muscle types evaluated under such circumstances, as it is well-known that the quality of each muscle differs as a result of their inherent differences.

The aim of the present study was thus to investigate the effects of two housing system types (caged versus penned) on the physiochemical and sensorial meat quality of a variety of important locomotive muscles from the hind leg together with the *longissimus thoracis et lumborum* muscle, as differing levels of movement/activity within each of the systems could influence these parameters through differing degrees of muscle development and fat deposition. It was hypothesised that the increase in physical activities able to be performed in pens could impact the sizes of these locomotive muscles, thereby affecting traits such as yield, colour, shear force, and will increase muscle protein concentration whilst lowering intramuscular fat (IMF) deposition. The result of these changes in muscle traits could have consequences on the sensory quality of rabbit meat, due to their relationships with texture, flavour, and aroma, especially the IMF concentration and its fatty acid profile.

2. Material and methods

The experiment was conducted at the Demonstration and Experimental Centre of the Faculty of Agrobiology, Food, and Natural Resources (Czech University of Life Sciences Prague) in the Czech Republic. All experimental procedures were in agreement with the Directive 2010/63/EU, revising Directive 86/609/EEC, regarding the protection of animals used for scientific purposes. Ethical clearance was granted by the Ethical Committee for Animal Experimentation at the Czech University of Life Sciences Prague (approval document number 10/2020).

2.1. Animals and experimental design

A total of 135 rabbits of the commercial Hyplus cross-bred line (PS 19 × PS 40), of both sexes (50% males and 50% females), were weaned at 36 d of age and randomly divided into two experimental groups according to housing system type. The experimental protocol closely follows that of Krunt et al. (2021). Sixty rabbits (balanced for sex across housing types) were kept in groups of three rabbits per cage (90 × 50 × 45 cm; 0.15 m^2 per rabbit), in a total of 20 cages ($n = 60$; 30 males, 30 females) alternating a sex ratio of 1 male:2 females and 2 males:1 female in each cage. The cages were constructed from a 3 mm diameter galvanized wire mesh floor and wire mesh walls. The remaining rabbits ($n = 75$; 38 males and 37 females) were randomly placed into one of three pens (25 rabbits/pen; 0.15 m^2 per rabbit); pen 1 contained 12 males and 13 females, pen 2: 13 males and 12 females, and pen 3 had 13 males and 12 females. The pens were constructed from a plastic mesh floor (10 mm slat width, 10 mm slot width, and 50% perforation) and wire mesh walls. The facilities were temperature- and humidity-controlled (18 to 21 °C, and 65–70% relative humidity) and the daily light period used was 12 h. Rabbits were fed ad libitum with a pelleted fattening diet (Table 1 and Table 2) and had ad libitum access to water via nipple drinkers.

Table 1
Ingredient and chemical composition of pelleted diet (%) according to Krunt et al. (2021).

Ingredient	%
Alfalfa meal	30
Sunflower meal	17
Wheat bran	23.5
Sugar beet pulp	4
Oats	13
Barley	8
Rapeseed oil	2
Vitamin-mineral premix	0.5
Monocalcium phosphate	0.5
Limestone	1
Salt	0.5
Chemical composition	
Dry matter	88.6
Crude protein	14.5
NDF	29.4
ADF	18.3
ADL	3.9
Ether extract	3.0
Starch	18.7
Ash	6.6
Ca	0.996
P	0.649
Mg	0.308
Digestible energy (MJ/kg)	10.2

Included per kg of diet: vitamin A, 12,000 IU; vitamin D3, 1500 IU; vitamin E, 50 mg; vitamin K3, 1 mg; vitamin B1, 2 mg; vitamin B2, 6 mg; vitamin B6, 3 mg; niacinamide, 20 mg; Ca-pantothenate, 10 mg; folic acid, 0.5 mg; vitamin B12, 0.02 mg; choline chloride, 500 mg; Co, 0.3 mg; Cu, 8 mg; Fe, 27 mg; I, 0.8 mg; Mn, 19 mg; Zn, 44 mg; Se, 0.07 mg. As provided by the compound feed manufacturer.
NDF, neutral detergent fibre; ADF, acid detergent fibre; ADL, acid detergent lignin.

2.2. Growth and slaughter performance

The rabbits were individually weighed once a week. Feed intake was recorded daily per cage/pen, from weaning until slaughter, for calculation of the feed conversion ratio. At 80 days of age, 28 rabbits were randomly selected (around the mean live weight for the respective housing system treatments) from the housing systems (balanced for sex) and slaughtered at commercial slaughterhouse. Slaughter and carcass dissection performance was evaluated according to the standards of the World Rabbit Science Association (Blasco & Ouhayoun, 1996). The rabbits were exsanguinated, skinned, and eviscerated (genitals, bladder, gastrointestinal tract), and the distal segment of the legs were removed. The remaining carcass (head, liver, kidneys, perirenal and scapular fat, thymus, trachea, oesophagus, lungs, and heart) were weighed to determine the hot carcass (HC) weight, and determine the dressing percentage relative to the live slaughter weight. After 1 h of suspension from the tendon calcaneus of both legs, the carcasses were chilled at 4 °C for 24 h, and then weighed again to determine the chilled carcass (CC) weight and calculate carcass cooler shrinkage (CCS). To determine the reference carcass (RC) weight for each individual carcass (according to Blasco & Ouhayoun, 1996), the head and internal organs were removed. The kidney fat, inguinal fat, and scapular fat was dissected from the carcass and weighed, to calculate its yield as a percentage of the RC. The carcasses were then sectioned by cutting between the 7th and 8th *thoracis vertebrae,* and between 6th and 7th *lumbar vertebrae*, to yield the fore, mid, and hind parts, which were then weighed separately, and their yield was calculated as a percentage of the RC weight. The hind leg (thigh) was removed from the hind part of the RC (Blasco & Ouhayoun, 1996), weighed (yield indicated as percentage of RC), and further dissected to remove the thigh muscle, which was also weighed, and its

Table 2
Fatty acid profile (% of fatty acid) of the rabbit diet fed to grower rabbits under two different housing systems.

Parameter	%
SFA	
12:0	0.04
14:0	0.15
15:0	0.06
16:0	10.04
17:0	0.07
18:0	1.75
Other SFA	0.62
Total SFA	12.77
MUFA	
14:1	0.01
16:1	0.29
18:1 n-9	37.95
18:1 n-7	1.52
20:1 n-9	0.81
Other MUFA	0.03
Total MUFA	40.85
PUFA	
18:2 n-6	36.76
18:3 n-3	9.00
20:2 n-6	0.06
20:3 n-6	0.01
20:4 n-6	0.04
20:5 n-3	0.17
22:5 n-3	0.03
22:6 n-3	nd
Other PUFA	0.12
Total PUFA	46.85

SFA, saturated fatty acids; MUFA, mono-unsaturated fatty acids; PUFA, polyunsaturated fatty acids; nd, not detected.

yield was calculated as a percentage of the RC.

2.3. Physical analyses

After cooling for 24 h at 4 °C, carcasses were deboned, and the right carcass side *longissimus thoracis* (LT), *longissimus lumborum* (LL), *biceps femoris* (BF), *semitendinosus muscle* (ST), and *quadriceps femoris* (QF) were removed, weighed, and their individual yields were determined as a percentage of the CC side weight. Muscle pH was then determined in each individual muscle (InoLab pH 730 set with automatic temperature adjustment, WTW, Weilheim, Germany). Meat cut surface colour (L*, lightness; a*, redness; b*, yellowness) was also measured at 24 h *post-mortem* (3 replicates per sample), on cut surfaces of steaks from each muscle after a 30 min blooming period, using a Minolta CM-700d spectrophotometer (8 mm aperture diameter size; 0% UV; D-65/10° illuminant/observer angle; Konica Minolta, Osaka, Japan). Samples were then cut from each muscle to determine cooking loss percentages, by weighing the samples, placing them within a plastic bag into a pre-heated water bath (80 °C) until an internal temperature of 75 °C (measured by a thermometer probe; AD14TH, Ama-Digit, Kreuzwertheim, Germany). The samples were allocated at random to one of four cooking batches. The samples were then removed, cooled to room temperature, blotted, and weighed again. From these samples, five cores (10 × 10 mm) were cut, parallel with the muscle fibres. The samples were placed into a Instron Universal Texture Analyzer 3365 (Instron, Canton, MA, USA) fitted with a standard Warner-Bratzler blade, and the peak shear force was recorded per sample at a crosshead speed of 100 mm/min and averaged across the five cores (sheared perpendicular to the direction of the muscle fibres).

2.4. Chemical analyses

The meat samples from the left hind leg were homogenised and frozen at −20 °C until analysis. The moisture content of the meat samples was determined by oven drying (105 °C) to a constant weight, whereafter samples were pulverized and used for crude protein (Kjeltec 2400, FOSS Tecator AB, Höganäs, Sweden), crude fat (Soxtec Avanti 2055 System, FOSS Tecator AB, Höganäs, Sweden), and ash determination (6 h at 550 °C). After fat extraction, the samples were used for determining the hydroxyproline content using a spectrophotometer (Varian Cary 50 Probe, Mulgrave, Australia) following the methodology of Bergman and Loxley (1963). The total collagen content was then calculated by multiplying the hydroxyproline content by a factor of 7.25 and expressing it as g/kg muscle. The energy value (kJ/100 g muscle) was then calculated as 16.75 x protein content +37.68 × fat content, according to the method described by Simonová et al., 2010).

2.5. Fatty acid analyses

Both the diet and meat samples from the hind legs intended for fatty acid (FA) analyses were analysed by gas chromatography following the extraction of total lipids as described by Volek, Bureš, and Uhlířová (2018). The FA proportions (g/100 g of FAs determined) were expressed as percentages of the total area of injected methyl esters. The content of FA (mg/100 g muscle tissue) was determined with nonadecanoic acid as an internal standard. Alkaline *trans*-methylation of fatty acid was done according to Raes, De Smet, Balcaen, Claeys, and Demeyer (2003). Chromatograph HP 6890 (Agilent Technologies, Santa Clara, CA, USA) with a programmed 60 m DB − 23 capillary column (150–230 °C) and a flame-ionization detector (FID) was used for performing the gas chromatography of methyl esters. An Agilent autosampler was used for performing split injections. Fatty acids were determined by retention times contrasted with standards. A known amount of internal standards was added to each sample - the standards were PUFA 1, PUFA 2, PUFA 3 and 37 Component Fame Mix (Supelco, Bellefonte, PA, USA). These were utilized to set up the calibration curve, with the equation:

$$A_i/A_{istd} = k_i \times (C_i/C_{istd}) + b$$

where A_i is analyte signal area of i, A_{istd} is internal standard signal area, C_i is concentration of the analyte i, C_{istd} is concentration of internal standard, k_i is response factor and b is constant.

Calculations of atherogenic (AI) and thrombogenic (TI) indices were performed according to Ulbricht and Southgate (1991), peroxidability index was calculated following the formula of Arakawa and Sagai (1986):

$$AI = [C12:0 + (4 \times C14:0) + C16:0] \Big/ \Big[\sum \text{MUFA} + \sum \text{n}-6\ \text{PUFA} + \sum \text{n}-3\ \text{PUFA}\Big]$$

$$TI = [C14:0 + C16:0 + C18:0] \Big/ \Big[\Big(0.5 \times \sum \text{MUFA}\Big) + \Big(0.5 \times \sum \text{n}-6\ \text{PUFA}\Big) + \Big(3 \times \sum \text{n}-3\ \text{PUFA}\Big) + \Big(\sum \text{n}-3\ \text{PUFA} \Big/ \sum \text{n}-6\ \text{PUFA}\Big)\Big]$$

$$PI = (\%\text{monoenoic} \times 0.025) + (\%\text{dienoic} \times 1) + (\%\text{trienoic} \times 2) + (\%\text{tetraenoic} \times 4) + (\%\text{pentaenoic} \times 6) + (\%\text{hexaenoic} \times 8)$$

2.6. Descriptive sensory analyses

Samples of the left *longissimus lumborum* (LL) muscles of all animals and were vacuum-packed and refrigerated at 4 °C for a further 24 h (total of 48 h from slaughter). The whole muscles were grilled on a preheated (200 °C) double glass/ceramic plate grill (VCR 6 L TL, Fiamma, Aveiro, Portugal) until an internal sample temperature of

70 °C, which was determined by a digital temperature probe (AD14TH, Ama-Digit, Kreuzwertheim, Germany). Muscle samples were cut into 15 mm-long rectangles, omitting the outer edges that had contact with the grill, placed in sealed glass containers (labelled with a randomized code), and stored at +50 °C until evaluation (~1 h).

Descriptory sensory evaluation was performed by six trained panellists (ISO 8586, 2012) in individual cubicles (ISO 8589, 2007) with red lightning, according to a linear unstructured continuous 100 mm scale for each of the eight descriptors, defined according to Volek et al. (2018). Two training sessions were performed before the analyses, using rabbit meat samples. The analysis was performed during two sessions within one day and included in a total of 28 samples evaluated in 14 sets, presented to panellists in randomized orders to avoid first-order carry-over effects and possible effects of the order of presentation. Panellists were provided with water and bread between samples. Each set had one sample from each housing system type and were from animals of the same sex.

2.7. Statistical analyses

Statistical analysis was performed using SAS statistical package (Version 9.4, SAS Institute, Cary, NC, USA). The measured variables were firstly tested for normality using the Shapiro-Wilk test (procedure UNIVARIATE), and for homogeneity of variance with Levene's test (procedure GLM). A mixed linear model was used (procedure MIXED), and parameters were estimated by the restricted maximum likelihood (REML) method. The evaluation of the growth parameters included the data from all 135 rabbits, whilst that of the slaughter and meat quality only included the 28 selected rabbits. The model for growth traits, carcass traits, physical, chemical and fatty acid data used the fixed effect of *housing system* and random effect of *pen/cage* and *animal*. The effect of *sex* was initially included as a fixed effect, without significant effects on the results, and thereafter was included rather as a random effect as sex was not the focus of this study.

The statistical model for growth, carcass traits, physical, chemical, and fatty acid data was:

$$Y_{ijkl} = \mu + H_i + p_j + a_{k+}\ e_{ijkl}$$

where:Y_{ijkl} = observed variableμ = overall meanH_i = fixed effect of housing system (i = 1, 2)p_j = random effect of pen/cage (j = 1–3/20)a_k = random effect of animal (k = 1–28)e_{ijkl} = random error

For the analyses of cooking loss and shear force, *cooking batch* was also accounted for as a random variable.

The model equation for cooking loss and shear force was:

$$Y_{ijkl} = \mu + H_i + a_j + b_k + e_{ijkl}$$

where:Y_{ijkl} = observed variableμ = overall meanH_i = fixed effect of housing system (i = 1, 2)a_j = random effect of animal (j = 1–28)b_k = random effect of batch (k = 1–4)e_{ijkl} = random error

The model used to evaluate sensory characteristics included the fixed effect of *housing system* and random effect of *gender* of the animals, *session*, and *assessor*. The model equation for sensory analysis was:

$$Y_{ijklm} = \mu + H_i + g_j + s_k + a_l + e_{ijklm}$$

where:Y_{ijklm} = observed variableμ = overall meanH_i = fixed effect of housing system (i = 1, 2)g_j = random effect of gender of animal (j = 1, 2) s_k = random effect of session (k = 1, 2)a_l = random effect of assessor (l = 1–7)e_{ijklm} = random error

The data are presented as the least squares means (LSM) and their respective standard errors (SEM).

Table 3
Growth performance according to housing system.

Parameter	Housing system		SEM	*P*-value
	Pen	Cage		
Live weight 36 d (g)	919	898	12	0.401
Live weight 80 d (g)	2791[b]	3023[a]	32	0.005
Period 36–80 d				
Body weight gain (g/d)	39.8[b]	46.5[a]	0.6	0.001
Feed intake (g/d)	138[b]	147[a]	4.98	0.039
Feed conversion ratio	3.30	3.13	0.08	0.561

[ab]Means in the same row with different superscript letters differ significantly at 0.05.
SEM, standard error of the mean (n = 25 rabbits per each of the 3 pens; 3 rabbits per each of the 20 cages).

3. Results

3.1. Growth performance characteristics

Table 3 shows the growth performance of the rabbits during the trial. The live weight at 80 days of age, body weight gain, and feed intake were significantly higher for the rabbits housed in cages compared to rabbits from pens.

3.2. Carcass characteristics

The results of carcass characteristics are displayed in Table 4. Differences were found in cooler carcass shrinkage, dressing percentage, thigh (% RC), and thigh muscle (% RC) yields. Significantly higher values of cooler carcass shrinkage were reported for rabbits from pens compared to rabbits from cages. The dressing percentage was also higher in pen-housed rabbits (+ 1.5%) than cage-housed rabbits. The thigh yields were significantly greater (+ 0.7%) in rabbits from pens than rabbits from cages. However, the thigh muscle yield was greater in caged rabbits than in pen rabbits (+ 0.6%).

3.3. Physical attributes of selected muscles

Differences between pen-housed and cage-housed rabbits were found for the physical attributes of their muscles (Table 5). Specifically, a lower a* colour value was found in the LT of penned rabbits compared with caged rabbits. On the other hand, rabbits from cages had meat with

Table 4
The effect of housing system on carcass characteristics of rabbits at the age of 80 days (28 rabbits per treatment group, gender ratio of 1:1).

Parameter	Housing system		SEM	*P*-value
	Pen	Cage		
Slaughter weight (g)	2866	2946	30.05	0.179
Hot carcass (g)	1745	1742	20.06	0.930
Chilled carcass (g)	1713	1717	19.09	0.917
Reference carcass (g)	1408	1404	16.50	0.910
Cooler carcass shrinkage (%)	1.85[a]	1.41[b]	0.07	≤0.001
Dressing percentage (%)	59.8[a]	58.3[b]	0.37	0.044
Fore part (% RC)	42.1	41.9	0.28	0.731
Mid part (% RC)	18.2	18.1	0.28	0.866
Hind part (% RC)	36.6	36.9	0.19	0.998
Thigh (% RC)	17.4[a]	16.7[b]	0.12	0.001
Thigh muscle (% RC)	13.6[b]	14.2[a]	0.13	0.032
Kidney fat (% RC)	2.1	2.07	0.14	0.912
Inguinal fat (% RC)	0.45	0.35	0.06	0.449
Scapular fat (% RC)	0.54	0.49	0.05	0.623
Total dissectible fat (% RC)	3.08	2.92	0.20	0.680

[ab]Means in the same row with different superscript letters differ significantly at 0.05.
SEM, standard error of the mean; RC, reference carcass.

Table 5
Effect of housing system on physical attributes of *longissimus thoracis*, *longissimus lumborum*, *biceps femoris*, *semitendinosus*, *and quadriceps femoris* muscles of rabbits at the age of 80 days (28 rabbits per treatment group, gender ratio of 1:1).

Parameter	Housing system		SEM	*P*-value
	Pen	Cage		
Longissimus thoracis				
pH_u	5.86	5.85	0.028	0.929
Colour *L** (lightness)	58.44	56.68	0.890	0.174
Colour *a** (redness)	-2.24[a]	-1.48[b]	0.174	0.005
Colour *b** (yellowness)	8.17	8.42	0.355	0.605
Drip loss (%)	1.56[b]	2.20[a]	0.200	0.032
WBSF (N)	24.41	24.23	1.152	0.918
Cooking loss (%)	27.27	27.53	1.380	0.894
Longissimus lumborum				
pH_u	5.77	5.82	0.039	0.327
Colour *L** (lightness)	58.31	59.19	1.360	0.465
Colour *a** (redness)	−1.53	−1.44	0.355	0.863
Colour *b** (yellowness)	8.15[b]	9.58[a]	0.497	0.050
Drip loss (%)	2.91	3.05	0.531	0.816
WBSF (N)	33.14	30.51	1.706	0.203
Cooking loss (%)	28.37	28.17	1.445	0.923
Biceps femoris				
pH_u	5.91	5.95	0.032	0.346
Colour *L** (lightness)	58.12	60.99	1.418	0.165
Colour *a** (redness)	−1.56	−0.99	0.368	0.283
Colour *b** (yellowness)	8.49	9.71	0.616	0.173
Drip loss (%)	0.74	0.56	0.093	0.188
WBSF (N)	28.39	27.16	0.913	0.353
Cooking loss (%)	26.12	26.65	0.736	0.606
Semitendinosus				
pH_u	6.05	6.07	0.045	0.667
Colour *L** (lightness)	55.50	55.24	1.092	0.868
Colour *a**(redness)	0.89	1.30	0.586	0.627
Colour *b**(yellowness)	7.81	7.54	0.771	0.802
Drip loss (%)	1.91	1.69	0.305	0.617
WBSF (N)	25.74	24.99	1.366	0.628
Cooking loss (%)	28.61	29.53	0.816	0.431
Quadriceps femoris				
pH_u	6.10	6.05	0.042	0.378
Colour *L** (lightness)	50.13	50.59	0.988	0.745
Colour *a** (redness)	3.61[a]	1.89[b]	0.798	0.016
Colour *b** (yellowness)	12.66[a]	10.98[b]	0.665	0.045
Drip loss (%)	1.01	0.82	0.151	0.356
WBSF (N)	29.56	27.17	1.887	0.194
Cooking loss (%)	27.29	27.67	0.913	0.771

[ab]Means in the same row with different superscript letters differ significantly at 0.05.
SEM, standard error of the mean; WBSF, Warner-Bratzler shear force.

higher drip losses than rabbits from pens. Rabbits from cages had higher LL b* colour values than rabbits from pens. The QF muscle differed in colour between the treatments, resulting in higher a* and b* colour values in the muscles from rabbits grown in pens compared to rabbits in cages. No significant influence of housing system on the physical attributes of the BF and ST muscles was found.

3.4. Fatty acid profile, including indices of human health

The contents of selected fatty acids and indices related to human health are shown in Table 6. Regarding the saturated fatty acids, a significantly higher content of C17:0 fatty acid was found in the meat from caged rabbits. For MUFAs, 18:1 n-9 and 20:1 n-9 also had higher concentrations in the meat from caged rabbits. Thus, the total MUFA concentrations were higher in the meat from cage-housed rabbits compared to that from pen-housed rabbits. The 20:3 n-6, 20:4 n-6, 22:5 n-3 concentrations were higher in the meat from penned rabbits.

Table 6
Fatty acid profile (mg/100 g of muscle from the left hind leg) and indices related to human health of rabbits at the age of 80 days (28 rabbits per treatment group, gender ratio of 1:1).

Parameter	Housing system		SEM	*P*-value
	Pen	Cage		
SFA				
12:0	2.36	3.01	0.27	0.230
14:0	22.98	30.97	2.36	0.092
15:0	5.92	7.42	0.44	0.091
16:0	311.5	380.9	19.16	0.070
17:0	6.21[b]	8.99[a]	0.60	0.022
18:0	99.01	115.5	5.06	0.100
Other SFA	4.13	5.30	0.33	0.080
Total SFA	452.1	552.1	27.80	0.071
MUFA				
14:1	2.90	5.94	0.83	0.071
16:1	41.00	58.94	5.06	0.080
18:1 n-9	281.2[b]	389.4[a]	24.07	0.023
18:1 n-7	19.74	22.90	1.13	0.171
20:1 n-9	5.64[b]	7.28[a]	0.42	0.050
Other MUFA	3.22	4.10	0.29	0.130
Total MUFA	353.7[b]	488.6[a]	31.12	0.021
PUFA				
18:2 n-6	288.3	315.5	12.75	0.290
18:3 n-3	31.39	38.21	2.17	0.121
20:2 n-6	4.31	4.67	0.19	0.353
20:3 n-6	7.10[a]	5.46[b]	0.24	≤0.001
20:4 n-6	54.18[a]	41.42[b]	1.86	≤0.001
20:5 n-3	2.23	2.04	0.07	0.141
22:4 n-6	5.49	5.16	0.15	0.272
22:5 n-3	10.88[a]	9.26[b]	0.36	0.020
22:6 n-3	2.36	2.07	0.08	0.080
Other PUFA	5.03[b]	5.94[a]	0.23	0.040
Total PUFA	411.3	429.7	15.72	0.571
n-3	48.78	53.49	2.33	0.323
n-6	362	375.3	13.46	0.630
PUFA n-6/PUFA n-3	7.47	7.13	0.13	0.181
Total fatty acids	1217	1470	71.51	0.084
Health indices				
AI	0.53	0.54	0.01	0.222
TI	0.86	0.89	0.01	0.231
PI	60.71[a]	49.22[b]	1.84	≤0.001

[ab]Means in the same row with different superscript letters differ significantly at 0.05.
SEM, standard error of the mean; SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; AI, atherogenic index; TI, thrombogenic index; PI, peroxidability index.

Table 7
The effect of the housing system on the chemical composition of the left hind leg meat of rabbits at the age of 80 days (28 rabbits per treatment group, gender ratio of 1:1).

Parameter	Housing system		SEM	*P*-value
	Pen	Cage		
Dry matter (g/kg)	254.9	254.1	0.85	0.652
Moisture (g/kg)	745.1	745.9	0.86	0.654
Fat (g/kg)	20.04	20.18	0.65	0.920
Protein (g/kg)	204.6	206.3	0.61	0.172
Ash (g/kg)	11.71[a]	11.34[b]	0.07	0.008
Hydroxyproline (g/kg)	1.32	1.29	0.02	0.561
Collagen R (g/kg)	4.36	4.18	0.09	0.353
Energy value (MJ/kg)	4.21	4.19	0.03	0.671

[ab]Means in the same row with different superscript letters differ significantly at 0.05.
SEM, standard error of the mean.

However, the other PUFAs were higher in the meat from caged rabbits, and as a result, the housing system did not influence the total PUFA concentrations. Regarding the calculated indices, the peroxidability index was greater for the meat from pen-housed rabbits.

3.5. Chemical composition of the hind leg

The hind leg meat's chemical composition (Table 7) was not affected by the housing system, except the ash content, which had a significantly higher concentration within the meat from penned rabbits.

3.6. Weight and muscle yields of selected muscles

Table 8 shows no significant differences between the housing systems regarding the weights and yields of the BF, ST, or QF muscles. The LL and LT muscle weights and yields (as a percentage of the fore part of the carcass) were influenced by the housing system, with caged rabbits having higher yields. No further effects were seen for the weights and yields of the BF, ST, or QF muscles.

3.7. Organoleptic properties of longissimus lumborum muscle

Table 9 displays the results of organoleptic properties of the grilled LL muscle, as affected by housing system. Housing system had no influence on the organoleptic properties of the grilled LL muscle.

4. Discussion

The growth performance of rabbits in different housing systems depends on the specifications of the system itself, from the types of materials utilized to the space available for certain behaviours (i.e., moving, resting, feeding), and other unique rearing conditions that the system provides (i.e., health and/or social stress). For example, a caged rabbit system typically provides less space for locomotion, and increases feed intake, resulting in higher weight gains compared to pen systems (Maertens & Van Herck, 2000). In addition, placing more rabbits within one pen can stimulate a higher degree of social interactions, despite utilizing the same stocking rate as a caged system. As a result of these social interactions, rabbit movement is likely to be further increased, as rabbits compete for resources (such as food and space) as well as hierarchical rank, while simply exhibiting more social interactions than a housing system with fewer individuals in one cage/pen.

Increased movement (Krunt et al., 2021) and/or competition for feed (Xiccato et al., 2013) can affect carcass traits and meat quality, through differences in muscle development and fat deposition. For example, the present study reports higher cooler carcass shrinkage in pen-housed rabbits compared to cage-housed, which has been explained by different growth rates connected to the differing energy expenditures of the animals as a result of higher movement rates under pen-housing (Dal Bosco, Castellini, & Bernardini, 2000). Thus, while caged rabbits have inferior dressing out percentages (DPs) and fatness, their carcases have less cooler shrinkage as a consequence, which would affect saleable carcass yields. The higher DP of penned rabbits in the present study could be associated with greater thigh yields (Table 4) due to muscle development (size) and/or development of their bones (density) (Matics et al., 2018; Krunt et al. (2021). The greater development of the hind part of the carcass - or greater thigh yield (% RC) - of rabbits with more opportunities for physical activity (such as penned rabbits) has also been reported by other studies investigated housing systems which allow for different degrees of physical activity (Combes et al., 2010; Krunt et al., 2021; Matics et al., 2018). In the present study, the individually examined muscles did not differ in their percentage yields, with the exception of the LL (primarily a so-called postural muscle), which had a slightly higher yield under caged conditions. Combes et al. (2010) reported higher LL and ST muscles weights in favour of rabbits from cages compared to pens, likely due to their greater slaughter weights, as caged rabbits were heavier than penned rabbits, reiterating the need to account for slaughter weight for yield comparison purposes.

When the effect of housing was examined on the selected individual muscles, differences were mostly found in colour, with higher value of redness (a^*) in the LT muscle and higher yellowness (b^*) in the LL muscle from rabbits in cages. On the other hand, redness (a^*) and yellowness (b^*) of the QF muscle were higher in pen-housed rabbit compared to cage-housed rabbits. When considering the effect of housing on the colour of the QF muscle in the present study, these results may be explained by the conclusion of Gondret, Hernandez, Remignon, and Combes (2009) who found that the increased redness (a^*) and yellowness (b^*) of the BF muscle were the consequence of the modified oxidative metabolism and myofiber types (rich in mitochondria and myoglobin) of this muscle, linked to higher animal movement in collective rearing systems. According to Ouhayoun (1998), increased movement affects muscle fibre type (and size), which can increase the proportion of so-called "red" muscle fibres compared to "white" muscle fibres, which differ in their mitochondria or myoglobin content, and can affect the colour of the meat. In addition, Gondret, Ramouz, Fernandez,

Table 8
Effect of housing system on muscle yields of *longissimus thoracis*, *longissimus lumborum*, *biceps femoris*, *semitendinosus*, *and quadriceps femoris* muscles of rabbits at the age of 80 days (28 rabbits per treatment group, gender ratio of 1:1).

Parameter	Housing system		SEM	*P*-value
	Pen	Cage		
Longissimus thoracis				
Weight of muscle (g)	20.33	18.20	2.155	0.068
Muscle yields (% of fore part)	6.13[b]	6.89[a]	0.202	0.050
Longissimus lumborum				
Weight of muscle (g)	65.56	62.79	2.171	0.375
Muscle yields (% of mid part)	48.96[b]	51.53[a]	0.494	0.007
Biceps femoris				
Weight of muscle (g)	37.27	37.67	0.994	0.777
Muscle yields (% of thigh)	19.62	18.73	0.271	0.112
Muscle yields (% of hind part)	7.31	7.19	0.101	0.533
Semitendinosus				
Weight of muscle (g)	10.48	10.34	0.562	0.776
Muscle yields (% of thigh)	5.39	5.28	0.121	0.645
Muscle yields (% of hind part)	2.00	2.03	0.045	0.809
Quadriceps femoris				
Weight of muscle (g)	28.74	26.81	1.788	0.061
Muscle yields (% of thigh)	14.00	14.50	0.300	0.412
Muscle yields (% of hind part)	5.22	5.56	0.110	0.141

[ab]Means in the same row with different superscript letters differ significantly at 0.05.
SEM, standard error of the mean.

Table 9
Effect of housing system on organoleptic properties of *longissimus lumborum* muscle of rabbits at the age of 80 days (28 rabbits per treatment group, gender ratio of 1:1).

Parameter	Housing system		SEM	*P*-value
	Pen	Cage		
Aroma intensity	65.9	65.5	4.12	0.872
Rabbit aroma intensity	63.4	60.1	2.78	0.188
Tenderness	60.4	61.2	3.46	0.823
Juiciness	60.0	56.9	2.87	0.329
Fibrosity	48.1	51.7	3.31	0.370
Flavour intensity	66.3	68.2	4.14	0.340
Rabbit flavour intensity	62.0	64.1	3.14	0.425
Chewiness	57.3	59.5	3.64	0.546

SEM, standard error of the mean.

and Combes (2004) also observed a larger number of type I fibres ("red") or II A ("white"), and an increased activity of citrate synthase and β-hydroxyl-Acyl-CoenzymeA dehydrogenase, in the *semimembranosus* and *biceps femoris* muscles of penned rabbits, when compared to conventionally reared counterparts. Increased meat redness is linked with a higher oxymyoglobin content, colour stability, and increased metmyoglobin-reducing activity (Neethling, Sigge, Hoffman, & Suman, 2018), which can prove advantageous for retail.

Based on the initial hypothesis, it was expected that rabbits from cage systems will have higher IMF content compared to penned rabbits; however, this result was not observed. Only the ash content of the meat differed, with a higher ash content in rabbits from pens than cages, which was similarly reported by Dalle Zotte et al. (2015). These findings can be further supported by Dalle Zotte et al. (2009), who explained that the hind leg meat is composed of several muscles with various levels of energy metabolism, and thus the rearing conditions can indeed affect the chemical composition of the meat. The amount of physical activity may indirectly influence the fatty acid profile. The deposition of PUFAs increase slower than that of SFAs and MUFAs, resulting in a decrease in the relative proportion of PUFAs (De Smet, Raes, & Demeyer, 2004). In response to physical activity, Sutherland, Woodhouse, and Heyworth (1981) showed a decrease in 18:1 and increase in 18:2 n-6 fatty acids in the adipose tissue of humans, likely due to the reduced activity of stearoyl-CoA desaturase. In animals, chronic exercise is connected to a decrease in MUFA content (Nikolaidis & Mougios, 2004), which correlates with the results of the present study, where rabbits from pens had significantly higher MUFA content than rabbits in cages, likely due to (jumping, running, escaping agonistic social interactions, etc.). According to Dalle Zotte et al. (2015), it can be assumed that differences in fatty acids between housing systems (pen vs. cage) are connected to the intramuscular fat content and the corresponding content of phospholipids, which are typical for their *n-6* and *n-3* content. The authors also draw attention to contradictions of comparing different muscles (hind leg vs. loin) due to oxidative energy metabolism of hind leg and more glycolytic metabolism of the loin muscles.

According to the sensory analysis, the housing systems did not significantly affect any of the observed organoleptic properties, despite observed effects on the meat fatty acid profile. As a typical sensory analysis presents only the cooked meat to the panellist, the fresh meat colour is not assessed. The major influencers of rabbit meat sensory properties are usually the breed (Fadare & Arogbo, 2015), age (Gondret, Juin, Mourot, & Bonneau, 1998), or nutrition (Volek et al., 2018). Thus, if rabbits are to be reared under penned conditions to higher slaughter weights/physiological development stages (84 days +) the combined effects of increasing age (in terms of sexual dimorphism, and the effects on agonistic behaviours on activity and stress) and differing degrees of muscle fibre trait development on physiochemical and sensory meat quality traits should not be ignored.

5. Conclusions

Despite differences in live performance parameters and carcass traits between rabbits from different housing systems, it appears that the housing system did not considerably influence the physiochemical and sensory meat quality of rabbits at studied age; however, these results should be interpreted with caution should rabbits be reared under penned conditions to higher slaughter weights/physiological development stages (84 days +). Considering the effect of housing system on meat fatty acid profiles, further research should focus on the oxidative stability and development of volatile compounds, which may also affect the point of purchase and repurchase of rabbit meat from different housing systems.

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Declaration of Competing Interest

The authors declare no conflict of interest.

Data availability

Data will be made available on request.

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animals

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Article

Effects of Genotype and Housing System on Rabbit Does' Aggressive Behaviors and Injuries in Smallholding Conditions

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Simple Summary: The housing of rabbit does in groups is nowadays a subject of study in the scientific literature due to the public's concerns about animals' welfare. Group housing of rabbits brings more social contacts and more space for manifesting their species–specific behaviours. However, group living has the potential to be more natural as rabbits live in colonies in nature, but the opposite is true. Problems with aggression among rabbit does are common in this type of housing. In addition, there are direct aggressive attacks by does towards kits from other mothers and this leads to economic losses due to higher kit mortality. There are various efforts to solve aggression among females, but most of them work with the implications of these procedures in intensive breeding, where the purchase prices of technology are very expensive. Therefore, this study deals with the solution to the given issue at the level of small farms, where the attention of the scientific sphere has been minimal.

Abstract: The objective of the study was to investigate the effects of housing (deep litter + concrete floor vs. deep litter + ground soil with the possibility to dig burrows), and genotype (Mecklenburg or Hyplus) on aggressive behaviour, social contacts, does' and kits' injuries, and progeny mortality. Twelve groups of six rabbit does (n = 72) were assigned to four treatments (two housing systems and two genotypes). Aggressive behaviour of does, number of injuries on does and kits, and postnatal kit mortality were recorded. The effects of housing and genotype were tested using multivariate GLMM Models. We found that the housing treatment in interaction with the genotype had a significant effect on aggressive behaviours in group housed does ($F_{3,12} = 14.34$, $p = 0.0003$), where the lowest incidence of aggression was in Mecklenburg does housed on ground soil. Reduced aggression was reflected in a lower number of injuries in does ($F_{3,68} = 10.51$, $p < 0.0001$), number of injuries in kits, and kit mortality ($F_{3,1} = 4.59$, $p < 0.0001$, $F_{3,54} = 43.94$, $p < 0.0001$). The results indicate that the proper combination of genotype and housing should be carefully considered for breeding to reduce aggression and injury in group housed does.

Keywords: aggression suppression; behaviour; burrows; injury; kits; smallholders

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1. Introduction

European wild rabbits (*Oryctolagus cuniculus*) live in colonies with their specific hierarchy, which is achieved by territorial behaviour. Rabbit groups usually consist of bucks two–three), females (two–nine), and their offspring. The hierarchy between males and

females manifests as linear. Females fight for breeding sites and males for territory and mates [1]. The dominant position among rabbits linearly changes in subsequent years according to their condition and is achieved by territorial and aggressive behaviour. Fights between the females are frequent at the beginning of breeding when strange animals are released together and gradually decrease after the social stabilisation of the group. Fighting is then rarer than in the beginning [2]. The trend of recent years is to bring the housing of rabbit does as close as possible to their natural behaviour (e.g., group housing). Regardless of the efforts of scientists to reduce aggressive behaviour between does kept on the farm, the aggressiveness of does towards each other is still a problem. Therefore, the females are usually single housed in a wire cage with their litter until weaning [3]. Aggression among rabbit does in group housing systems has been recently reported in several studies [4–6]. These studies investigated the influence of the group size, pen characteristics, or pen floor type on the behaviour and welfare of does. Typically, the aggression mainly took place during the first hour after grouping [4]. Scientific research has focused on two types of group housing systems (continuous and part-time) in recent years [7]. The authors summarised that continuous group housing resulted in a high number of aggressive events and injuries in does, which is incompatible with animal welfare. They see the potential in part-time group housing; however, the final procedure which could be applied to farms has not been developed yet. Szendrő et al. [8] reviewed the use of hiding places, which reduced the number of does with skin injuries and were shown to be promising in group-housed systems [9]. Although many studies have looked at the effect of the housing system, there are very few that take into account the effect of rabbit genotype; however, there were some indicia (more aggressive Dutch rabbits, compared to New Zealand ones) which revealed this possibility to be relevant [10].

There are nearly 161,000 registered backyard farms and 4500 commercial farms with 180 million rabbits for meat production in the EU [11]. Backyard farming is divided into three classes: small, medium, and large farms. The farmers usually follow family traditions in breeding animals, doing as their parents or grandparents did. Owners of medium and large backyard farms are usually interested in expanding their knowledge and improving the quality of animals' lives as well as farm production. Intensive rabbit production is typical in the EU, particularly for countries such as Italy, France, Spain, and Hungary. On the other hand, small and medium farms are spread all around the world, predominating in South America and Asia. For these farms, scientific knowledge is essential [12]. Moreover, there are backyard farms, which have limited resources, and farming systems are on the "low input" level in Africa. The housing systems are composed of local materials, and rabbit consumption usually takes place in the farmer´s house or local market [13]. Typically, native, or coloured breeds are used in small or medium size farms for meat production. Rabbit does are bred there naturally when the doe is released into the buck´s cage for mating [12]. Based on the literature research, we designed an alternative housing system for group-housed does for smallholders. The rearing system was based on the possibility of making burrows (does are housed on the ground consisting of a layer of soil with deep littering manifesting their species-specific activities, e.g., digging). We have compared this housing system with deep litter housing (both with a male presence) with the concrete ground, where the does could not make burrows. Moreover, we used Hyplus genotype rabbits that are commercially used, and the original breed Mecklenburg checked rabbits for comparison.

We hypothesised that rabbit does with the possibility of digging burrows will show aggressive behaviour towards other females less often, will have fewer skin injuries, fewer kits with skin injuries, and lower kit mortality than does from deep litter housing. Furthermore, Mecklenburg checked rabbit does with the possibility of making burrows will show aggressive behaviour towards other females less often, will have fewer skin injuries and fewer kits with skin injuries, and have lower kit mortality than Hyplus females.

2. Materials and Methods

2.1. Animals and Housing

The study was piloted at the Czech University of Life Sciences Prague using 72 24-week-old rabbit does: slow-growing does of Mecklenburg rabbits (MC) and commercial hybrid Hyplus, where all housing conditions were simulated. The observations took place from May until July 2021. Rabbits were fed a commercial pelleted diet (Sehnoutek a synové, Czech Republic), water (provided by nipple drinkers), gnawing material (wooden sticks), and hay were available ad libitum. From 10 weeks of age, female rabbits were housed individually (deep litter with access to hay) in the open-air system. At 24 weeks of age, does were randomly assigned to four treatments depending on the floor type—concrete with deep litter (DEEP) or soil (the depth of the soil was ad libitum for rabbits, so the females could dig however they wanted) with deep litter (DIG)—and genotype (MC or Hyplus). The does were divided according to genotype (36 does per genotype) and according to the floor type (DEEP or DIG). Each treatment consisted of three replications of six rabbit does according to genotype and housing system. The does were weighed at the beginning and the end of the experiment (after kindling). The average weight of MC does at the beginning of the experiment was 4561 g ± 169 SE and 4503 g ± 180 SE in Hyplus does. The weight of MC does was 4434 g ± 225 SE and 4208 g ± 165 SE in Hyplus does after kindling on average. After four days of socialisation, a male of the same genotype was released into the group. As specified by [14], the male was present for ten days in each group and then he was removed. After these ten days, all does were detected as gravid by palpation (kits were born during two days). The housing scheme can be seen in Figure 1. Pens had the tops open, and the average size was 3.5 × 2.0 m with a minimum surface area of 1.6 m^2 per doe. A reverse "U" shape platform (1 × 0.6 m), elevated platform (0.4 m above the ground), and nest boxes (0.3 × 0.4 × 0.3 m) were counted in. The nest boxes were in two positions accessible for the does: either on the ground or in elevated areas. The number of nest boxes was always two more (8 in total) than the number of does in the pen to prevent unwanted aggressive behaviour during the choosing of the nest. In the DIG group, the nest boxes were placed 10 cm below the ground for better stability. Elevated platforms and resting areas were made of wood with no perforation. Moreover, in the central part of the pen, there was a platform with the reverse "U" shape, which also served as a hiding spot. Each housing system was equipped with hiding tubes lying on the floor (1 × 0.25 m). The entire area of the pen (concrete × soil) was completely covered (15 cm depth) with deep straw litter (chopped straw with an average length of 0.1 m), which was removed every week. Housing systems were placed outdoors and protected by netting against intruders.

Growing rabbits, after weaning were visually checked every week for control of their health status until the end of the slaughter (not a part of the present paper).

2.2. Behavioural Observation and Analyses

All pens were video recorded with colour infrared cameras (Sikur systems, Frýdek-Místek, Czech Republic). The period of observation had five parts: the initial period of the group formation, when the male was added 1 week before kindling; 14 days after kindling; and 35 days after kindling (contact with growing kits). Videos were recorded twice in 30-min-long intervals (day and night, from 10:00 to 10:30 and 22:00 to 22:30, resp.) during each period. Aggressive behaviour against does (does were marked by different colours) was observed according to [15] in 1-min intervals by the one-zero sampling method (i.e., the behaviour being present at any time during the 1-min interval) for 30-min periods. The sums of values obtained from the reproductive period were counted for each behaviour and doe (marked by spray paint). The video clips were assessed by one trained researcher. Aggressive events were evaluated according to [16] as biting (gripping with the teeth); boxing (hitting with the front paws); chasing (aggressive following of another individual for at least three jumps); carousel-fights (rapid chasing around in one spot with the rear end of the opponent gripped between the teeth); attacking (aggressive running towards another female); and threatening (quick head movement toward another doe). These events were

counted together, and the sum of them was considered aggressive behaviour. "Sniffing" was considered as contact between two females who sniff each other without an aggressive ending to the event.

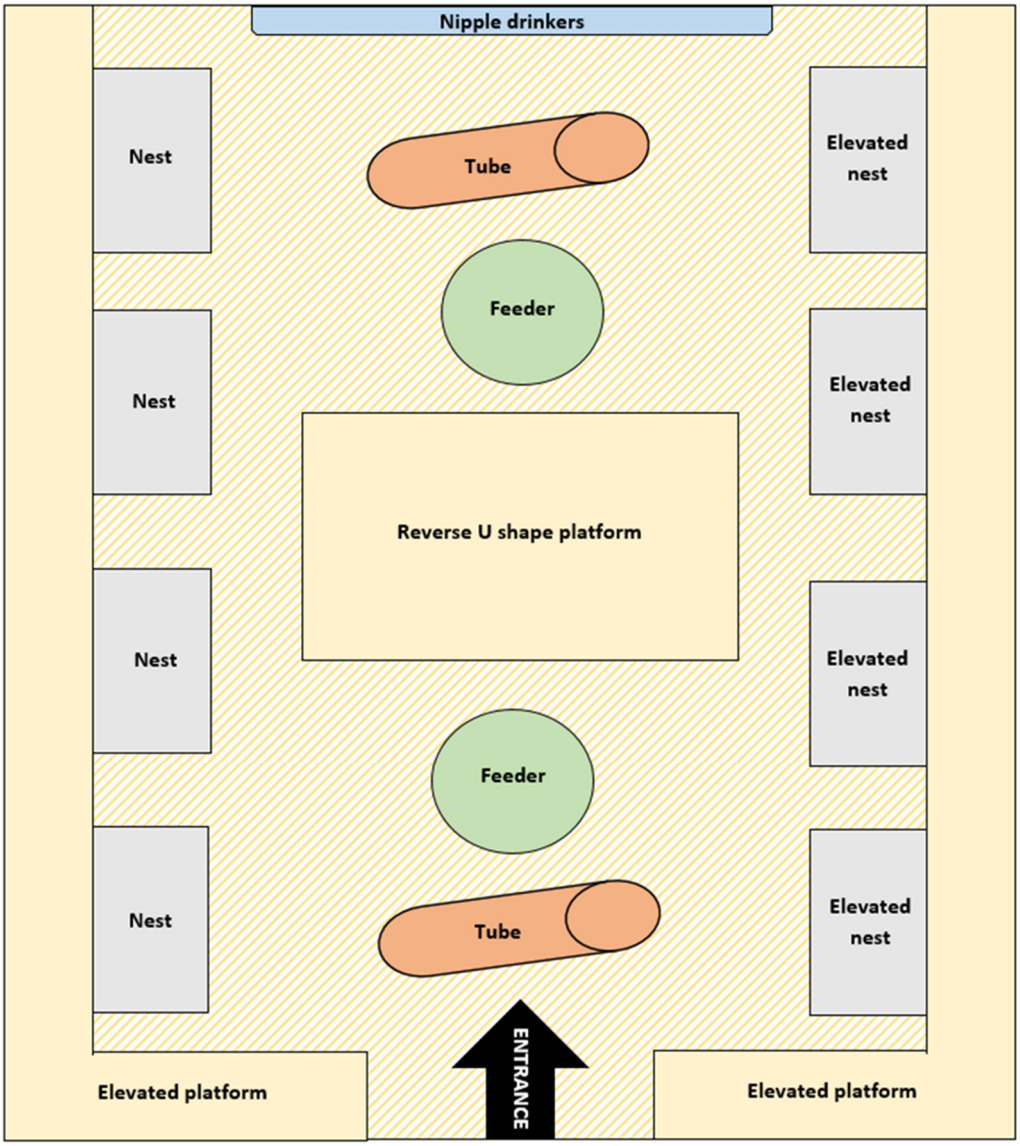

Figure 1. Housing system scheme (housing systems differed between groups by floor type).

2.3. Injury Scoring in Does

The does were individually checked for injuries before their release into the new group (day zero). Then, all does were scored three days after the group formation, three days after the male was added, three days after the male was removed from the group, twice (once a week) before kindling, and six times (once a week) after kindling until the kits were weaned. Each doe was carefully treated by an experienced researcher. Each female was captured and examined, old and healed injuries were excluded after the visual examination (fully or almost fully healed ones), and new ones were counted. The scoring schedule was conducted according to [9], where (0) were no skin injuries; (1) denoted small (<1 cm) superficial skin injuries (scratches), fewer than 6 in total; (2) denoted superficial skin injuries > 1 cm, deeper lesions in the connective tissue or more than five lesions score 1; (3) denoted very deep lesions in the muscle tissue (wounds) or more than five lesions as in score 2. We estimated the does' injuries by two variables: as a sum of scores; and by counting the number of injuries per doe without scoring. However, since these

two variables were highly correlated ($r = 0.94$, $p < 0.0001$), we used the latter variable for the analysis.

2.4. Injury Scoring in Kits

Kits were examined from 21 days of age until weaning once a week (three times in total). The sum of injuries in kits was counted for the observed periods. Kits usually did not receive wounds or deep bites (if they do, they were found dead). On their bodies were found in most cases little bites or scratches, so we considered counting the number of injuries on kits as reasonable and did not use the scale.

2.5. Postnatal Kits Mortality

Dead kits were recorded during the whole experimental period. Most bitten kits were found dead, so doe attacks were assumed to be the cause of death. Moreover, these kits had visible wounds on their body. In addition, to the total sum of dead kits we also added kits which were found dead in nests during first days after kindling, as the result of a fight between two does in the nest (visible on cameras).

2.6. Statistical Analyses

All data were initially tested for normality (Shapiro–Wilk test) and homogeneity of variances. Data validation was approached by calculating information on the validation and/or assurance quality procedure and output. Associations between dependent variables—aggressive behaviour towards other does (aggressive behaviour), does sniffing the other does within the pen (sniffing does), the quantity of skin injuries in does (doe injuries) and injuries in kits, and postnatal kit mortality (characteristics of the dependent variables in Table 1)—and independent variables (listed in Table 1) were tested using multivariate General Linear Mixed Models (GLMM, PROC MIXED in SAS 9.4). All GLMMs were designed for repeated measures (i.e., in SAS, where REPEATED = period; and SUBJECT = identity of the doe nested within the group) with the random effect (in SAS, RANDOM = group nested within housing).

Table 1. Countable and categorical variables available of moderating rabbit does aggressiveness by housing conditions and genotype.

Countable Variables	Mean	SE [1]	Min	Max	Unit
Aggressive behaviour	0.61	0.04	0	7	Number of cases
Sniffing does	1.18	0.07	0	15	Number of events
Doe injuries	112.92	2.59	0	330	Number of injuries
Kit´s injuries	46.17	4.01	3	93	Number of injuries
Kit natality	38	3.98	30	45	Number of kits born per group
Postnatal kit mortality	8.08	0.61	1	14	Number of dead kits per group
Doe's body weight	4531	6.52	4140	4960	g
Categorical variables		Levels			Description
Housing		2			DEEP [2]/DIG [3]
Genotype		2			Hyplus/MC [4]

[1] SE. standard error; [2] DEEP, deep litter housing; [3] DIG, digging housing system; [4] MC, Mecklenburg rabbits.

For the dependent variables, aggressive behaviour, sniffing does, and doe injuries, we constructed the models by first entering the interaction between housing and genotype, and then checking the model with the addition of the factors which could also affect the result (Table 1).

We could only measure the dependent variables, injured kits and postnatal kit mortality, during periods 4 and 5. Therefore, for these dependent variables, we removed the repeated measures design and analysed the GLMM with the interaction between genotype and housing and then checked the model with other possible factors from Table 1. Least-

squares means (LSMEANs) were computed here and thereafter for each class and, where appropriate, differences between classes were tested by *t*-test. We used a Tukey-Kramer adjustment for multiple comparisons. Associations between the dependent variable and fixed effects were estimated by fitting a random coefficient model using PROC MIXED as described by [17]. We calculated the predicted values of the dependent variables and plotted them against the fixed effects with a regression line. Statistical significance was based on $p < 0.05$.

3. Results

In total, we observed 21,600 one-min-long sampling intervals (300 for each doe) during which we registered 439 aggressive events: 143 in MC does, and 296 in Hyplus does. We further registered 847 events when a doe was sniffing another doe, 352 in MC does, and 495 sniffing events in Hyplus does. Overall, we had 212 kits from MC does and 244 kits from Hyplus does. In total, during the experiment, we counted 271 skin injuries on does, 96 on MC does, 175 on Hyplus does, and 554 skin injuries on kits, 177 in MC, and 377 in Hyplus kits. We found 97 dead kits, 25 MC kits, and 72 Hyplus kits.

3.1. Number of Attacks among Does

In partial agreement with the advanced hypothesis, housing and genotype had a significant effect on the number of attacks among does (Table 2), where MC does from DIG housing performed a smaller number of attacks towards other does compared to MC does from DEEP housing ($p = 0.0082$), as shown in Figure 2. There were also significant differences when comparing does from the same housing treatment but different genotypes. MC does from DIG housing had fewer attacks compared to Hyplus does from DIG housing ($p = 0.0002$), and MC does from DEEP housing had less attacks compared to Hyplus does from DEEP housing ($p = 0.0128$). However, there was no significant difference in the number of attacks between Hyplus does from DIG and Hyplus does from DEEP housing.

Table 2. GLMM results (fixed effects, degrees of freedom, *F*-Value, and probability) of moderating rabbit does aggressiveness by housing conditions, and genotype.

Fixed Effect	Num DF	Den DF	*F*-Value	*p*-Value
Dependent variable: aggressive behaviour				
Housing*genotype	3	12	14.34	0.0003
Body weight	1	62.6	4.93	0.0301
Dependent variable: sniffing				
Housing*genotype	3	12	2.67	0.0946
Dependent variable: injuries in does				
Housing*genotype	3	68	20.40	<0.0001
Dependent variable: injuries in kits				
Housing*genotype	3	1	4.59	<0.0001
Litter size	1	60	8.71	0.0045
Dependent variable: postnatal kit mortality				
Housing*genotype	3	54	43.94	<0.0001
Number of injuries in kits (genotype)	2	54	9.28	0.0003

*, It is a sign for interaction.

In addition, body weight had a significant effect on the number of attacks among does (Table 2), with heavier females attacking significantly more often than lighter females (Figure 2).

3.2. Doe Sniffing the Other Does

Differences in housing and genotype did not reach statistical significance for sniffing among the does within the pen (Table 2).

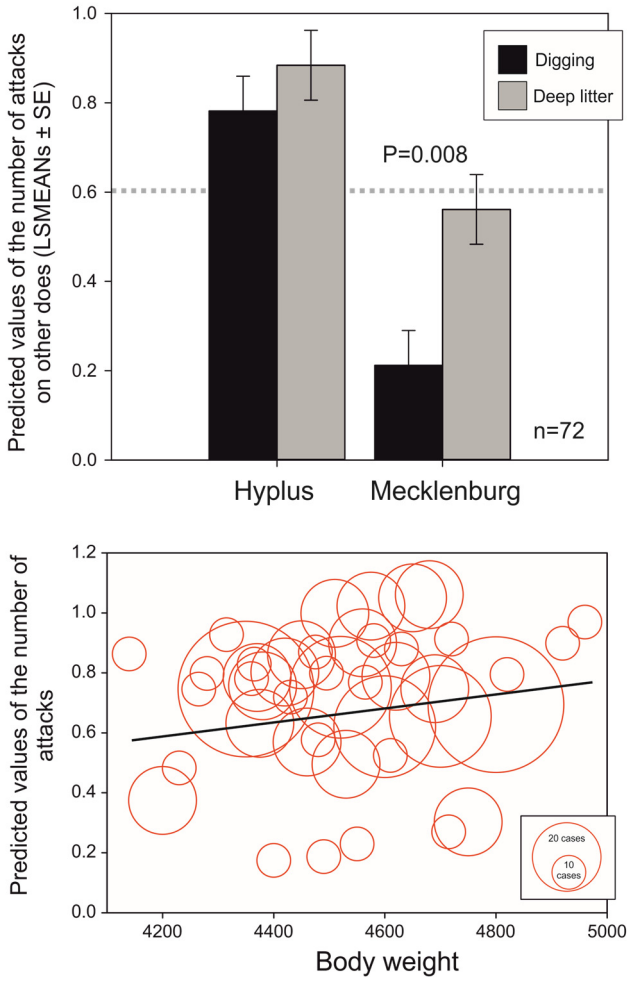

Figure 2. Predicted values of the number of attacks on other does according to housing (**top**) for Hyplus (left column) and Mecklenburg rabbits (right column); LSMEANs ± SE, n = 72 for each column, plotted against the body weight (**bottom**).

3.3. Number of Skin Injuries of Does

The number of skin injuries and the severity of injuries were strongly correlated (r = 0.942). Reduced aggression was reflected in a lower number of injuries in does (Table 2, Figure 3), where MC does from DIG housing had fewer skin injuries compared to MC does from DEEP housing (p = 0.0092). There were also significant differences when comparing does from the DIG housing treatment but different genotypes. MC does from DIG housing had fewer injuries than Hyplus does from DIG housing ($p < 0.0001$). However, there were no significant differences in the number of injuries in MC does from DEEP housing compared to Hyplus does from DEEP housing and no differences in the number injuries between Hyplus does from DIG housing and Hyplus does from DEEP housing. Body weight also had no effect on the number of skin injuries.

3.4. Number of Injuries in Kits

The housing treatment in interaction with the genotype had a significant effect on the injuries of kits (Table 2, Figure 4), where MC kits from DIG housing had fewer injuries than MC kits from DEEP housing ($p < 0.0001$) and Hyplus kits from DIG housing had fewer injuries than Hyplus kits form DEEP housing ($p < 0.0001$). Furthermore, the number of injuries of kits was also significantly affected by the size of the litter (Table 2); the more kits were in the litter, the more injuries the kits had (Figure 4).

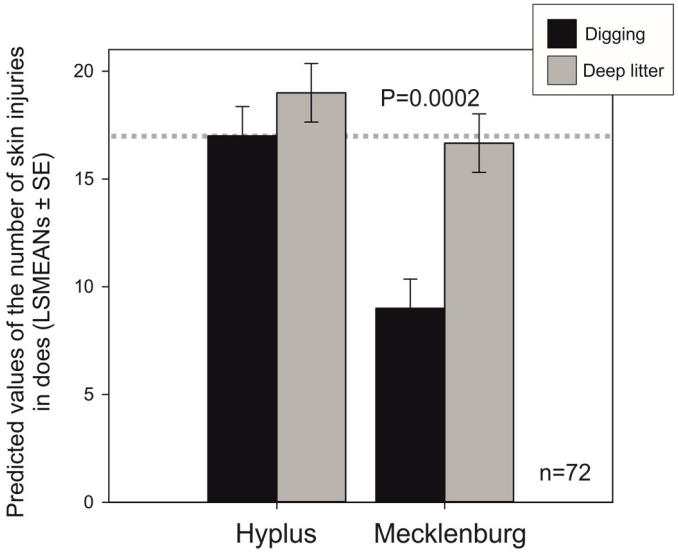

Figure 3. Predicted values of the number of skin injuries in does according to housing for Hyplus (left column) and Mecklenburg rabbits (right column); LSMEANs ± SE, n = 72 for each column.

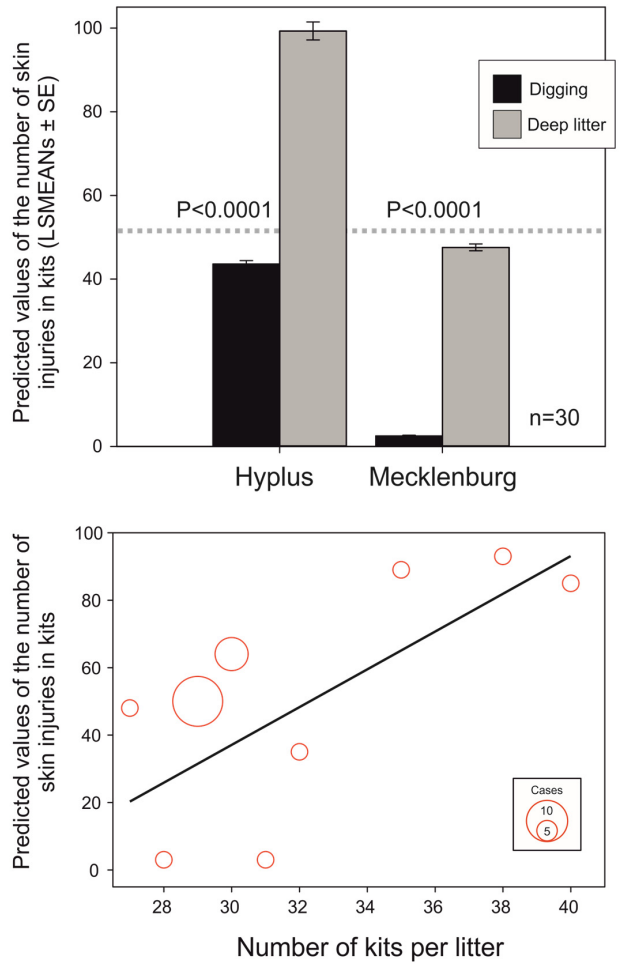

Figure 4. Predicted values of the number of skin injuries in kits according to housing (**top**) for Hyplus (left column) and Mecklenburg rabbits (right column); LSMEANs ± SE, n = 30 for each column, plotted against the number of kits per litter (**bottom**).

3.5. Postnatal Kit Mortality

Housing and genotype also had a significant effect on postnatal kit mortality (genotype nested within housing $F_{3,54} = 43.94$, $p < 0.0001$). Kit mortality was lower in MC kits in DIG housing than in MC kits in DEEP housing ($p < 0.0001$) as seen in Figure 5. There were also significant differences when comparing the mortality of kits from the DIG housing treatment with different genotypes. MC kits from DIG housing had lower mortality than Hyplus kits from DIG housing ($p < 0.0001$). However, there were no significant differences in the mortality of MC kits from DEEP housing compared to Hyplus kits from DEEP housing. The mortality rate was also no different for Hyplus kits from DIG housing compared to Hyplus kits from DEEP housing. The number of injuries had a significant effect on mortality (injuries nested within genotype $F_{2,54} = 9.28$, $p = 0.0003$). In both genotypes, the higher the number of kits injured, the higher the kit mortality (Figure 5).

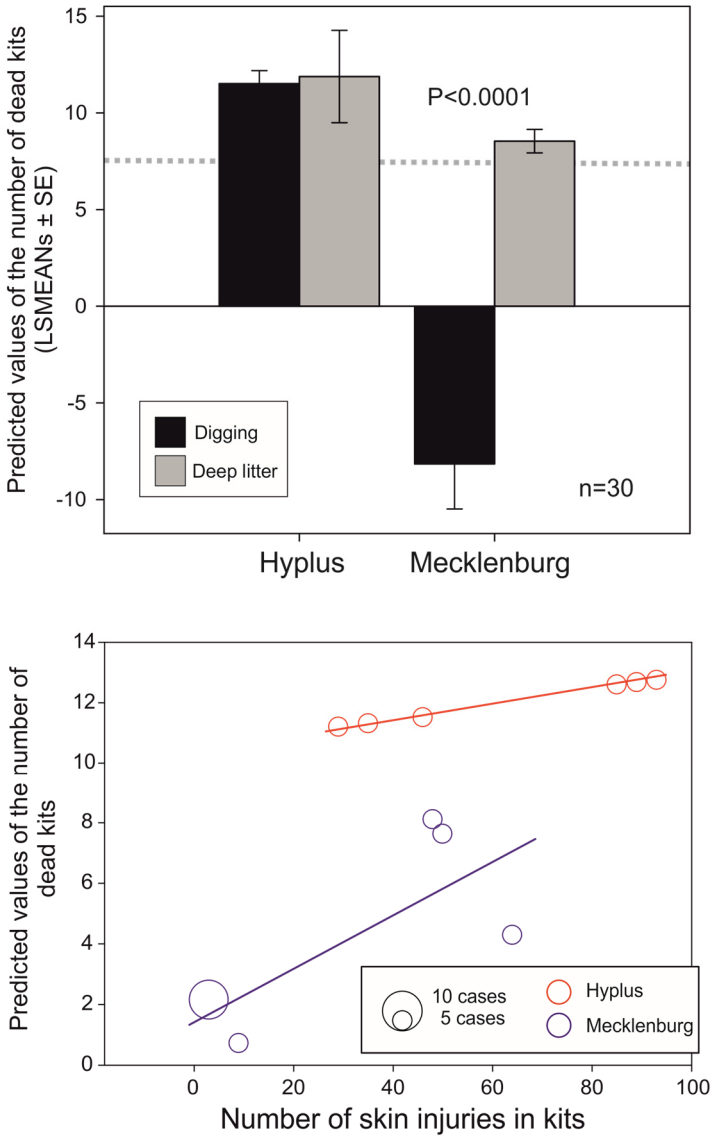

Figure 5. Predicted values of the number of dead kits according to housing (**top**) for Hyplus (left column) and Mecklenburg rabbits (right column); LSMEANs ± SE, n = 30 for each column, plotted against the number of skin injuries in kits (**bottom**) for Hyplus (red) and Mecklenburg rabbits (blue).

4. Discussion

The results of the present study showed that the housing system and genotype had a significant effect on the aggressive behaviour of rabbit does. Generally, when the does were housed in groups (continuous or part-time), they manifested a high frequency of aggressive events before the hierarchy of the group was established [18–20]. This trend was also observed in the present study, where both genotypes showed a high level of aggressive behaviour in the first observed period. Several studies tried to solve the problem of aggression with different regrouping schedules [21] or multiple strategies, when platforms, plastic pipes, hiding places, straws, dark corridors, or sprayed odours were tested, and the results were rather unconvincing, as reviewed by [7]. These findings were used in our study, where the deep litter systems were equipped with appropriate (previously tested) elements (such as hiding pipes, gnawing sticks, and shelters), and the digging systems were enriched with the same elements and the possibility of creating a rabbits' warrens under the ground and doing related activities.

In our study, the commercial Hyplus was more aggressive in general than the MC genotype. However, their inter-individual social behaviour, reflected in sniffing, did not differ. It is likely that Hyplus does are more aggressive than MC does because they are selected primarily for productive parameters [22], regardless of behaviour. Contrarily, on small farms or at breeders (these breeders typically use the original or local breeds on their farms), it is very common to eliminate aggressive rabbits from breeding, as the rabbits are frequently used as pets [23]. Another possible explanation for our results could be hidden in the hypothesis which postulates that some individuals possess genetic variants enhancing their vulnerability to environmental adversity. This was reported by [24] in humans, where the longitudinal data of several hundred people were used, resulting in relatively common variants of the dopamine receptor gene and the serotonin transporter gene interacting with social conditions to predict aggression in a manner consonant with the differential susceptibility perspective. The authors reported that in case of adverse environmental conditions, the individuals with these genetic variants manifested more aggression than other genotypes. Additionally, the effect of genotype on aggressive behaviour was previously confirmed by [25], who observed differences in expressing this behaviour among various mouse strains. This is also well-known in chickens, where white-feathered birds are more aggressive than red ones [26], which is generally correlated with some candidate genes [27]. However, these studies considered that genetics could be the key determinant of aggressive behaviour; studies related to humans are in this way more detailed. Moreover, we found a higher number of attacks directed from heavier to lighter does. The information about the effect of body weight on aggressive behaviour is lacking in the scientific literature. Only [28] reported no correlation between body weight and rank order in growing rabbits from 4 to 12 weeks of age.

Aggression is not reflected just in hurting other does, but also in injured or even killed kits. Ref. [29] reported that wild rabbit does tolerate their kits but demonstrate aggression against other offspring. In addition, aggression can exceed into infanticide due to the limited number of burrows. These events naturally lead to increased kit mortality and therefore economic losses. In the present paper, a higher incidence of mortality was observed in the Hyplus genotype, which was more aggressive compared to MC does, as was previously mentioned.

The housing system influenced aggression in the less aggressive MC genotype only. Hyplus does maintain aggression at a high level during the whole reproductive period. It has to be emphasised, however, that the does' injuries had never been severe enough to lead to inflammatory conditions. Still, we recorded more injured does and kits in DEEP systems compared to DIG systems. Surprisingly, the injuries led to mortality only among the MC genotype. The greater part of dead kits among the Hyplus genotype occurred during the first days after leaving of the nest. The aggression is mostly noted at the start of the reproductive season and after parturition, when rabbit does are intolerant of each other [30]. Problems with aggression are therefore reported with respect to both strategies

of housing rabbit does. Ref. [6] tested four individual modules and a common area, where the rabbit does were mixed after 18 days after kindling, resulting in 50% injured does. Furthermore, [31] observed groups of five does in a part-time system with results showing 34% injured does four days after group formation and 53% injured does at litter weaning. In addition, during lactation, kits were the subject of aggressive attacks from other does. These attacks can be then the cause of death as assumed in [32], who observed a significant effect of grouping rabbit does at different times post-partum. In our study, the MC genotype housed in DIG systems seemed to be more compatible than Hyplus. Hyplus may manifest more aggressive behaviour and related injuries, or deaths could occur due to their "higher distance" from wild rabbits as these lines are bred specifically for high production. These findings could be explained by the study of [33], who compared domestic and wild rabbits and found a higher number of aggressive attacks among wild rabbits than their domestic counterparts. Based on the results, it seems that the genetic selection of reproductive traits could also affect the maternal behaviour of rabbit does, which, in selected strain, could increase aggressive behaviour in protecting the nest and the litter. In the present study, the MC does are calmer; they do not look for fights. On the contrary, Hyplus fights more for hierarchy and may need a bigger space for their nests to eliminate attacks from other does or kits.

5. Conclusions

The results show that the choice of genotype and housing system is crucial with respect to aggressive interactions among rabbit does. Aggression results in injuries among rabbit does and their kits. Fights among does can sometimes occur in the nest and new-born kits will subsequently die. Kits that are seriously injured by other rabbit does later die. The MC genotype, as the original medium-heavy breed, is less aggressive than the commercial hybrid we tested and can be recommended for smallholder breeding conditions. For these farmers, it is also important to choose a suitable housing system; according to our results, the possibility of digging should not be missing, and housing systems should accommodate the building of nests of greater complexity. In addition, although we did not observe any health problems in does or their kits, the potential risk of coccidiosis infection is present when using deep litter instead of a slatted or wire floor. It would be interesting to test the genotypes of rabbits typically used for meat production on small farms in future research. Moreover, other studies should focus on the effect of genetic selection on the maternal behaviour of rabbit does.

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Institutional Review Board Statement: The welfare of the rabbits was carefully considered during the whole experiment. The animals were not subjected to pain, suffering, distress, or lasting harm. Feed and water were provided ad libitum. The whole study was carried out in harmony with the guidelines of Act No. 246/1992, which directs on protection against animal cruelty. The study was approved by the ethics committee of the Czech University of Life Sciences Prague.

Informed Consent Statement: Not applicable.

Data Availability Statement: The data are available upon reasonable request.

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Original Paper

Housing of rabbit does: reproductive performance, health and bone quality

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The objective of the present paper was to evaluate the health status reflected in haematological and biochemical traits of the blood, reproductive performance and bone quality of rabbit does on first parturition housed in several housing systems with or without mirrors. The animals were randomly allocated into 6 experimental groups: pens with soil covered by deep litter with the possibility of digging burrows (A), pens with soil covered by deep litter with the possibility of digging burrows enriched with a mirror (B), pens with plastic slatted floor with elevated resting area (C), pens with plastic slatted floor with elevated resting area enriched with a mirror (D), combi-park system (E) and combi-park system enriched with a mirror (F). Litter size after birth and the number of weaned kits was found to be highest in group C, D, E and F compared to other groups. However, the biochemistry analyses showed every investigated trait as significantly affected by housing conditions except the UREA and CHOL, it has to be stated that values of the blood traits were in accordance with the reference values of rabbits. The effect of housing system on *tibia* and *femur* characteristics was found to be significant in fracture toughness, where the lowest value of this trait was detected in group E and F compared to other groups in both investigated bones. According to these findings, it is possible to recommend every tested housing system from a health point of view for farmers dealing with alternative housing systems.

Keywords: biochemistry, bones, haematology, housing system, reproductive performance

1 Introduction

Breeding rabbit does is now the subject of scientific research (Szendrő et al., 2019; Huang et al., 2021) due to welfare consequences of their housing. In farming systems, individual wire cages are mostly used, where the does are housed with their kits until weaning (Huang et al., 2021). During the last few years, the continuous and part-time collective housing of rabbit does was exposed to research resulting in non-acceptable conditions of rabbit does and their offspring (injuries in does or injured and dead kits) (Szendrő et al., 2019) or the health of the animals in general. For the description of the health status or for the prediction of disease or infection, the serum analysis is standardly used (Rehman et al., 2017). In addition, the decline in reproductive performance in collectively housed females is reflected in the number of pseudo-pregnancies or sharing the same nest box by more animals, which is usually followed by injuries in does and dead kits (Szendrő et al., 2019). To avoid these problems, part-time collective housing seems to be perspective (Zomeño et al., 2018). Several types of housing systems with different enriching elements were investigated ((e.g., hiding places; Rommers et al. (2014) or combi systems with removable walls; Dal Bosco et al. (2019)). Furthermore, the use of mirrors was investigated during the fattening period of growing rabbits resulting in a decreased incidence of stereotypical behaviour (Piller et al., 1999). When rabbits have the opportunity to choose, they always chose the part of the housing system, where the mirror was present (Dalle Zotte et al., 2009a). Another

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benefit of alternative housing usage is that rabbits have more space for movement and thus they have a higher fracture toughness of their bones compared to their counterparts from cages (Krunt et al., 2021).

The objective of the present paper was to evaluate the health status reflected in haematological and biochemical traits of the blood, bone quality and reproductive performance of rabbit does housed in several housing systems with or without mirrors. The overarching hypothesis was that the rabbit does housed in different housing conditions will show different blood profiles and different reproductive performances.

2 Material and methods

The present study was approved by the ethics committee of the Czech University of *Life Sciences* Prague. The whole study was carried out in harmony with the guidelines of Act No. 246/1992, which focuses on the protection of animals against cruelty.

2.1 Animals and husbandry

The study was conducted using 72 20-week-old rabbit does (Hyplus genotype) on first parturition. The animals were randomly allocated into 6 experimental groups according to housing system. The entire observation took place under natural conditions from June until September 2021. The rabbits were fed *ad libitum* by a hopper feeder with a standard fattening pelleted diet (17.80% CP, 9.9 Mj.kg^{-1} of digestible energy). Water was available *ad libitum* from nipple drinkers. The litter was supplemented *ad libitum* (groups A and B). Young rabbit does (11 weeks of age) were from weaning until the star of the experiment housed individually on plastic slatted floor, and at 20 weeks of age were divided into experimental groups (A, B, C, D, E, F) depending on the floor type. Every group consists of 6 animals. After one week, artificial insemination was performed. Each treatment consisted of 2 replications/6 rabbits.

Regarding the housing systems, pens with soil covered by deep litter with the possibility of digging burrows (A), pens with soil covered by deep litter with the possibility of digging burrows enriched with a mirror (B), pens with plastic slatted floor with elevated resting area (C), pens with plastic slatted floor with elevated resting area enriched with a mirror (D), combi-park system (E) and combi-park system enriched with a mirror (F) were used. The mirrors (50 × 120 cm) were installed permanently for the experimental period. Pens (A, B, C, D) had the top open and the average size was 2.5 × 3.0 m with a minimum surface area of 1.4 m^2 per doe after resting platforms and other enriching elements were counted in. Pens C and D were designed to have the same parameters as previously mentioned systems with two floors The floors were connected by an elevated step, which was used by rabbits to jump onto and then jump to the second floor. Under the second floor was a space divided into individual sections, which looked like a corridor. At the end of each corridor was a nest box. Each section was separated from the next one by a wire net wall to keep the rabbits in contact. The E and F systems were classic combi-park systems with removable walls designed for six rabbit does and their kits. All housing systems with rabbits were exposed to natural lighting and temperature, which was 16 : 8 L/D and 16 °C in average. Rabbit does were weighed at the start (21 weeks of age) and at the end of the experiment (31 weeks of age). Rabbit kits were weighed at birth and every week until weaning (35 days of age). At the end of the experiment, rabbit does were slaughtered (mechanically stunned) to obtain *tibia* and *femur* bones for analysing.

2.2 Haematological and biochemical blood sampling

Haematocrit (HCT), haemoglobin (HGB), erythrocytes (ERY), leukocytes (LEU), neutrophiles (NEU), lymphocytes (LYM), neutrophiles (NEU), monocytes (MO), eosinophiles (EOS), basophiles (BAS), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), plasma albumin (ALB), total protein (TP), urea (UREA), glucose (GLU), creatinine (CREA), globulin (GLOB), ALB/GLOB, alanine aminotransferase (ALT), alkaline phosphatase (ALP) and cholesterol (CHOL) contents were measured to indicate health status and body fitness of examined rabbits.

In the end of the experiment, blood samples from 4 animals (randomly selected) from each group and replication (48 samples) were taken from the ear vein. Collected samples were centrifuged at 700 × g for 20 min at 4 °C to obtain blood serum. Then the blood films were made using the slide method, previously described in Schalm et al. (1975). Determination of blood haemoglobin and haematocrit was done according to Dukes and Schwarte (1931). MCV, MCH and MCHC were calculated as described in Al-Daraji et al. (2008). Pappenheim May-Grunwald Giemsa stain was used for staining the blood films. ERY were determined by hemacytometer as well as NEU, MO, EOS and BAS. Three horizontal edge fields followed by two fields towards the centre were done to obtain a differential amount of LEU. Two more fields were lead in a horizontal direction and another two fields in a vertical direction to get the edge and obtain the crisscross shape with right angles. The rest of the blood serum was stored until the biochemical analysis, which was done using commercial kits (Erba Lachema, s. r. o., Czech Republic) on the automatic analyzer XL – 200 (Erba Lachema s. r. o., Czech Republic).

2.3 Bone quality analyses

The *tibia* and *femur* were frozen (-20 °C) after slaughter, for future analysing, in individual plastic bags. A day before the analyses, the bones were taken from the freezer and thawed overnight. Next, they were boiled for 15 min in 95 °C water, further stripped of flesh and dried at 25 °C for 24 h. Subsequently, the maximum shear force until initial structural failure (i.e., the breaking of the bone) was determined by a three-point flexure test using a Instron® Model 3342 (Instron, Norwood, Massachusetts, US) and the load rate was 12 mm.min^{-1}. The distance between the two fulcra points maintaining the bones was 45 and 38 mm. The bones were constantly oriented for testing with their natural convex shape downwards.

2.4 Statistical analyses

The statistical analyses were processed using the computer application SAS 9.4 (SAS Institute Inc. Cary. NC. USA). All the data were analysed using the General Linear Model:

$$Y_{ij} = \mu + HS_i + e_{ij}$$

where: Y_{ijk} – value of trait; μ – general mean; HS_i – effect of housing system (i = A–F); e_{ij} – random residual error

Housing system was considered as a fixed effect. Differences between means were determined by the Duncan's test. The value of $P \leq 0.05$ was considered significant for all measurements. All the data are expressed in tables as means.

3 Results and discussion

During the reproductive period no females died. In two females (one from group D and one from group E) were detected footpads. The initial weight of live females was between 4,413 and 4,613 g. The final live weight after weaning was between 4 643 and 4 986 g. The weights did not statistically differ among the groups in these two observation periods. Litter size after birth was found to be the highest in group C, D, E and F. The highest number of weaned kits was observed in the same groups as live born kits. Weaning weight of kits was found to be the highest in A, B, E and F groups. It has to be stated that does from group A and B had the lowest number of kits and their weaning weight was probably influenced by this factor. Typically, scientific literature compares group and individual housing with the conclusion of a decreased productive performance in group housed rabbit does compared to individually housed does (Maertens & Buijs, 2016; Dal Bosco et al., 2019). Maertens & Buijs (2016) reported a decline in the number of weaned kits and in their weaning weight in groups of females. Dal Bosco et al. (2019) found decreased litter sizes after birth and after weaning in group housed does compared to these from individual cages. In addition, the decline in reproductive performance in collectively housed females is reflected in the number of pseudo-pregnancies or sharing the same nest box by more animals, which is usually followed by injuries in does and dead kits (Szendrő et al., 2019). Sharing of the same nest box was observed also in the present study once in group D. We solved that problem by dividing little kits into two nest boxes, where females were separately housed with them for three days. Separated females still had contact with other members of the group through the wire walls. Then they were released and incorporated back into the group.

In general, the haematological and biochemical blood traits are used to indicate disease, infection or health condition (Rehman et al., 2017). The results of the effect of the housing system on haematological parameters of the rabbit does blood are displayed in Table 2. The investigated traits did not significantly differ among the groups. In a study by (Musco et al., 2019) it was found that rabbits, who were exposed to mirrors had higher values of RBC, MCV and MCH compared to their counterparts, who did not have mirrors in their housing system. The authors see a link between feed intake and growth rate,

Table 1 The effect of housing system on reproductive performance of rabbit does on first parturition

Trait	Housing system						*P*-value	SEM
	A	B	C	D	E	F		
LW of does 21 wks (g)	4,445	4,501	4,543	4,613	4,413	4,535	0.2634	45.486
LW of does 31 wks (g)	4,643	4,797	4,812	4,986	4,792	4,825	0.0561	41.553
Live born kits (n)	7.50^{b}	8.00^{b}	10.00^{a}	9.50^{a}	9.50^{a}	9.00^{a}	0.0001	0.1781
Weaned kits (n)	7.00^{b}	7.00^{b}	9.00^{a}	9.00^{a}	8.50^{a}	9.00^{a}	0.0001	0.1639
Weaning weight of kits (g)	972^{a}	985^{a}	872^{b}	866^{b}	967^{a}	960^{a}	0.0001	8.6247

A – pens with soil covered by deep litter with possibility of digging burrows; B – pens with soil covered by deep litter with possibility of digging burrows enriched by a mirror; C – pens with plastic slatted floor with elevated resting area; D – pens with plastic slatted floor with elevated resting area enriched by a mirror; E – combi-park system; F – combi-park system enriched by a mirror; LW – live weight; SEM – standard error of mean; values with significance of $P \leq 0.05$ were considered as significant

which is (when the growth rate declines) connected with feed utilization and reduced absorption of iron.

Furthermore, the biochemistry analyses showed (Table 3) every investigated trait as significantly affected by housing conditions except the UREA and CHOL. Rabbits from A and B group had the highest values of GLU, while the lowest values were found in C and D group. GLU is the main energy source (Gallenberger et al., 2012). Musco et al. (2019) observed the lowest values of GLU in rabbits, who were the most active. Unsurprisingly, our data indicates that rabbits from C and D had a high energy output thus they could jump from lower to upper floor, meanwhile the rabbits from digging systems obviously did not spend energy on movement as the previously mentioned rabbits. Additionally, according to measured values, we can state that rabbits were not exposed to stress, because values elevated above the optimum range indicate stress factors in rabbit's environment (Özkan et al., 2012). Moreover, CREA was significantly highest in B, D and F group, which are the groups with mirrors, and the lowest in A group. Musco et al. (2019) also found the highest values of CREA in mirrored groups compared to the rest of the groups. According to Bush (1991), the pre-renal failure is manifested by elevated in CREA Our results do not indicate any problems with rabbit does' kidneys. Also, measured TP was in the optimum range when the highest values were in A and C and the lowest values in B and E groups. It is well known that total protein and albumin are protein utilization indicators (Pavlík et al., 2007) and reflect the health condition of the animal (Marono et al., 2017). ALB values of group C, D and F were a bit above the optimum range, declared by Özkan et al. (2012). Significantly higher levels of GLB were observed in B, C, D and F groups than in other groups. The level of globulin in blood serum serves as an index immune reply. The higher globulin values or the higher ALB/GLB (in optimum range) the higher disease resistance and better immune response occurs in animals (Griminger, 1986). In the present paper, the highest value of ALB/GLB was detected for groups A and F and the lowest was in group B. The difference could be explained by some non-specific immune response initiated by environmental causes (El-Shafaei et al., 2016) thus the animals from group B had contact with the soil. Other studied parameters were ALT and ALP. These indicators are considered to reflect liver function similar to aspartate aminotransferase (Musco et al., 2019). Specifically, ALP serum levels come from the liver and bones and thus differ among the ages of animals and changes could be caused by bone growth (Kaneko et al., 1997). The obtained values in this study were found in the reference range, when ALT values were highest in groups A, B, D and F compared to the rest of the groups and ALP was detected as being higher in groups A and C compared to other groups. In addition, CHOL content did not significantly differ among the groups. However, in our study the presence of mirrors did not affect the CHOL content, Musco et al. (2019) referred to the highest amount of CHOL in mirrored groups of rabbits compared to these without mirrors.

Table 2 The effect of housing system on some haematological blood traits of rabbit does on first parturition

Trait	Housing system						*P*-value	SEM
	A	B	C	D	E	F		
HCT (%)	39.7	38.9	39.3	40.4	40.5	40.9	0.4242	0.580
HGB (g.l^{-1})	126	120	119	122	123	125	0.6041	2.451
ERY (10^{12}.l^{-1})	6.60	6.30	6.40	6.5	6.54	6.42	0.4782	0.064
LEU (10^{9}.l^{-1})	7.30	7.10	7.34	7.73	7.51	7.54	0.6410	0.324
NEU (10^{9}.l^{-1})	3.69	3.23	3.34	3.33	3.41	3.32	0.7641	0.201
LYM (10^{9}.l^{-1})	3.11	2.89	3.13	3.02	2.90	3.14	0.3678	0.086
MO (10^{9}.l^{-1})	0.730	0.840	0.738	0.748	0.761	0.745	0.6458	0.064
EOS (10^{9}.l^{-1})	0.312	0.291	0.300	0.333	0.294	0.289	0.5841	0.005
BAS (10^{9}.l^{-1})	0.239	0.211	0.242	0.220	0.232	0.236	0.8342	0.008
MCV (fl)	63.8	63.2	63.2	63.3	63.1	63.1	0.4132	0.194
MCH (pg)	19.6	19.6	19.3	19.9	19.6	19.4	0.6347	0.052
MCHC (g.l^{-1})	305	311	307	310	311	310	0.4246	1.102

A – pens with soil covered by deep litter with the possibility of digging burrows; B – pens with soil covered by deep litter with the possibility of digging burrows enriched by a mirror; C – pens with plastic slatted floor with elevated resting area; D – pens with plastic slatted floor with elevated resting area enriched by a mirror; E – combi-park system; F – combi-park system enriched by a mirror; HCT – hematocrit; HGB – hemoglobin; ERY – erythrocytes; LEU – leukocytes; NEU – neutrophiles; LYM – lymphocytes; MO – monocytes; EOS – eosinophiles; BAS – basophiles; MCV – mean corpuscular volume; MCHC – mean corpuscular hemoglobin concentration; SEM – standard error of mean; values with significance of $P \leq 0.05$ were considered as significant

Table 3 The effect of housing system on selected biochemical traits of rabbit does on first parturition

Trait	Housing system						*P*-value	SEM
	A	B	C	D	E	F		
GLU (mmol.l^{-1})	7.42^{a}	7.21^{a}	5.79^{c}	5.55^{c}	6.23^{b}	6.28^{b}	0.0135	0.301
CREA (µmol.l^{-1})	73.3^{c}	95.3^{a}	80.0^{bc}	94.5^{a}	86.5^{b}	93.24^{a}	0.0225	2.421
UREA (mmol.l^{-1})	8.38	8.50	8.33	8.43	7.62	7.324	0.2310	0.305
TP (g.l^{-1})	83.0^{a}	67.4^{c}	85.8^{a}	77.3^{b}	64.4^{c}	78.2^{b}	0.0001	1.421
ALB (g.l^{-1})	41.2^{b}	35.0^{c}	49.0^{a}	47.6^{a}	36.1^{bc}	47.1^{a}	0.0001	1.234
GLB (g.l^{-1})	28.3^{bc}	35.3^{a}	37.0^{a}	35.5^{a}	28.4^{c}	32.4^{a}	0.0001	0.749
ALB/GLB	1.45^{a}	0.99^{c}	1.32^{b}	1.34^{ab}	1.27^{b}	1.46^{a}	0.0002	0.062
ALT (UI)	88.0^{a}	91.0^{a}	76.0^{b}	97.5^{a}	69.7^{c}	95.5^{a}	0.0001	2.824
ALP (UI)	46.2^{a}	34.6^{b}	43.8^{a}	22.0^{cd}	33.0^{bc}	31.4^{bcd}	0.0001	2.159
CHOL (mmol.l^{-1})	1.32	1.32	1.25	1.30	1.13	1.29	0.0682	0.002

A – pens with soil covered by deep litter with the possibility of digging burrows; B – pens with soil covered by deep litter with the possibility of digging burrows enriched by a mirror; C – pens with plastic slatted floor with elevated resting area; D – pens with plastic slatted floor with elevated resting area enriched by a mirror; E – combi-park system; F – combi-park system enriched by a mirror; GLU – glucose; CREA – creatinine; TP – total protein; ALB – albumin; GLB – globulin; ALT – alanine transaminase; ALP – alkaline phosphatase; CHOL – cholesterol; SEM – standard error of mean; values with significance of $P \leq 0.05$ were considered as significant

Table 4 The effect of housing system on quality of *tibia* and *femur* bones in does on first parturition

Trait	Housing system						*P*-value	SEM
	A	B	C	D	E	F		
Tibia								
Length (mm)	109	110	108	110	112	110	0.7561	2.124
Width (mm)	8.3	8.42	8.15	8.24	8.16	8.21	0.547	0.214
Fracture toughness (N)	480^{a}	498^{a}	486^{a}	485^{a}	398^{b}	405^{b}	0.036	19.226
Femur								
Length (mm)	103	105	104	106	104	105	0.684	2.025
Width (mm)	8.71	8.68	8.59	8.64	8.62	8.67	0.267	0.242
Fracture toughness (N)	348^{a}	354^{a}	361^{a}	348^{a}	290^{b}	295^{b}	0.042	17.125

A – pens with soil covered by deep litter with the possibility of digging burrows; B – pens with soil covered by deep litter with the possibility of digging burrows enriched by a mirror; C – pens with plastic slatted floor with elevated resting area; D – pens with plastic slatted floor with elevated resting area enriched by a mirror; E – combi-park system; F – combi-park system enriched by a mirror; SEM – standard error of mean; values with significance of $P \leq 0.05$ were considered as significant

The effect of housing system on *tibia* and *femur* characteristics (Table 4) was found to be significant in fracture toughness, where the lowest value of this trait was detected in groups E and F compared to other groups in both investigated bones. The bone quality in rabbit does is marginally studied. Nevertheless, Buijs et al. (2014) published a paper, where they compared semi-group wire pen, semi-group plastic pen and individual cage system with results of significant differences among the groups in cortex thickness of *tibia* and *femur* bones. Rabbit does from individual cages had lower measured values than the other groups. They did not differ in the rest of the parameters (length, width, or breaking strength). The study of Krunt et al. (2021) compared collective pens with cages resulting in thinner bones in growing rabbits from cages compared to their counterparts. Both studies have common intersection-increased movement. The authors from the last cited study proved that bones of rabbits from pens contain more calcium and magnesium (*tibia* bone) or just magnesium (*femur* bone), which are crucial elements for determining bone quality and fracture toughness. Moreover, Dalle Zotte et al. (2009b) found, except the higher fracture toughness in penned rabbits compared to caged rabbits, that pen housed rabbits had heavier bones than those from cages.

4 Conclusions

Housing conditions significantly affected the number of live born and weaned kits and the weight of weaned young rabbits. Based on haematological and biochemical blood results, we can state that rabbit does were, during

the experimental period, healthy, and the obtained values were in the optimum range of physiological blood values for rabbits. According to these findings, it is possible to recommend every tested housing system from a health point of view. Moreover, fracture toughness was found to be the lowest in the combi-park system compared to the rest of the investigated systems, which is not the best option for dynamical movement of rabbit does. For future research, we can recommend focussing on stress hormone levels in similar group housing systems to obtain more detailed results and implement them into practise.

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4 Souhrnná diskuse

4.1 Produkční parametry a parametry jatečně upraveného těla ve vztahu k systému ustájení

Podmínky ustájení ovlivňují parametry růstu králíků ve výkrmu (Lambertini a kol., 2001; Princz a kol., 2009; Xiccato a kol., 2013; Matics a kol., 2018; Krunt a kol., 2021; 2022). Obecně platí, že rychlost růstu je nižší v systémech ustájení, které mají charakter výběhů, ohrad, nebo se jedná o různé modifikace boxů než u králíků chovaných v klecích po jednom nebo v malých skupinách (Lambertini a kol., 2001; Princz a kol., 2009; Xiccato a kol., 2013). Chov králíků ve výběhu je totitž logicky spojen s výkrmem větších skupin zvířat na společném prostoru (Krunt a kol., 2021). Parametry produkce, jako porážková hmotnost, přírůstek hmotnosti, příjem krmiva nebo konverze, jsou ovlivněny různými faktory, mezi které patří například chování zvířat, jež se napříč ustájeními liší. Například kompetice o krmivo (Xiccato a kol., 2013), vyšší příjem krmiva v klecích (Maertens a Van Herck, 2000), nebo zvýšená pohybová aktivita v ohradách (Krunt a kol., 2021) přispívají k rozdílnému stupni vývoje svalstva, a tedy rozdílné hmotnosti jednotlivých jatečných partií, případně zastoupení tuku v jatečném těle, a tedy ovlivnění hmotnosti zvířete.

Zvýšený pohyb (Krunt a kol., 2021) a/nebo konkurence o krmivo (Xiccato a kol., 2013) mohou ovlivnit charakteristiky masa a kvalitu masa prostřednictvím rozdílů ve vývoji svalů a ukládání tuku. Obecně lze konstatovat, že zvýšená míra pohybu, která je často spojena s nižším příjmem krmiva, generuje nižší jatečnou výtěžnost, nižší zastoupení tuku v těle a snižuje poměr masa ke kosti (Combes a kol., 2010). Jatečná výtěžnost je převážně ovlivněna právě příjmem krmiva. Králíci, kteří mají vyšší spotřebu krmiva, mají více plný trávicí trakt a jsou těžší při porážce (Trocino a kol., 2004). Systém ustájení má tedy největší vliv na jatečnou výtěžnost a rozvoj zadní části králičího těla/trupu, a to v souvislosti s pohybem zvířat. Králíci, kteří mají více prostoru k pohybu většinou vykazují vyšší hmotnost zadní části, vyšší osvalení stehen a vyšší hmotnost kostí stehen (Matics a kol., 2018; Krunt a kol., 2021). Ne vždy ale existují statistické rozdíly pro vývin zadní části mezi jednotlivými systémy (Krunt a kol., 2022). K vyšší hmotnosti stehen u králíků z výběhů v porovnání s těmi z klecí, přispívá i hmotnost kosti stehna. Na základě výsledků studie Krunta a kol. (2021) se ukázalo, že je hmotnost kosti systémem ustájení průkazně ovlivněna, a to pravděpodobně opět z důvodu vyššího pohybu ve výbězích. Pohyb a cvičení jsou jedny z faktorů, které ovlivňují hmotnost kosti ve spojitosti se zastoupením vápníku, fosforu a hořčíku (Clark son a Haymes, 1995). Například běh na

běžeckém pásu je jedním z forem cvičení pro krysy. Konkrétně jsou kosti stehenní a holenní podrobeny většímu mechanickému zatížení než bederní páteř (Iwamoto a kol., 2005). Navíc cvičení podporuje pozitivní vápenatou rovnováhu a zvyšuje hmotnost kostí (Yeh a kol., 1989), a tedy je prokázáno, že má pohyb vliv na kvalitu kostí.

4.2 Kvalita masa ve vztahu k systému ustájení

Kvalita masa je charakterizována mnoha parametry, mezi které patří parametry chemické, fyzikální, biochemické, senzorické, nutriční a jiné. Ty jsou pak charakterizovány dílčími parametry jako obsah tuku, bílkovin, pH, barva, obsah glykogenu, křehkost masa, výživová hodnota a další. S ohledem na výkrm králíků v různých systémech, hodnoty pH mají tendenci klesat s rostoucí velikostí skupiny (Combes a kol., 2010). Na hodnotu pH má vliv například agresivní chování zvířat, které podmiňuje adaptivní stresovou reakci ve svalu pro kontrolu většího množství volných radikálů vzniklých metabolicky. Čím nižší je pH, tím je také maso světlejší (Krunt a kol., 2020). To je způsobeno smrštěním kontraktilních vláken, která zvyšují rozptyl světla (Szendrő a Dalle Zotte, 2011). Barva masa je take ovlivněna pohybem, který zvyšuje energii metabolismu oxidativních svalů. Svaly s oxidativním metabolismem se nacházejí zpravidla na zadní končetině a je to například *biceps femoris* a typicky glykolytický metabolismus je u svalů hřbetu, jimiž je například *longissimus lumborum* (Dalle Zotte a kol., 2015). Hodnota pH těchto svalů se zvyšuje se snižující se hustotou osazení klece nebo výběhu (Preziuso a kol., 2009). Naopak pro hodnoty červenosti a žlutosti jsou napříč výzkumy rozdílné a není možné ve výsledcích, které o nich referují najít jasné schéma s ohledem na velikost skupiny (Szendrő a Dalle Zotte, 2011). Navíc, jak ukázala naše studie (Krunt a kol., 2022), je třeba referovat vždy o hodnotách L*a*b* vztažených ke konkrétnímu svalu s ohledem na jeho metabolismus, například *biceps femoris, semitendinosus* nebo *quadriceps femoris.* Z hlediska chemického složení masa, suma fyzické aktivity může nepřímo ovlivnit profil mastných kyselin. Ukládání PUFA probíhá pomaleji než u SFA a MUFA mastných kyselin, což vede k poklesu relativního podílu PUFA (De Smet a kol., 2004). V reakci na fyzickou aktivitu Sutherland a kol. (1981) objevili snížení 18:1 a zvýšení 18:2 n-6 mastných kyselin v tukové tkáni lidí, pravděpodobně v důsledku snížení aktivity stearoyl-CoA desaturasy. U zvířat je chronická fyzická aktivita spojena s poklesem obsahu MUFA (Nikolaidis a Mougios, 2004), což koresponduje s výsledky studie Krunt a kol. (2022), kde měli králíci z výběhu výrazně vyšší obsah MUFA než králíci v klecích, pravděpodobně kvůli aktivitám jako je skákání, běhání a úniky před agresivními sociálními interakcemi jiných zvířat. Podle Dalle Zotte a kol. (2015) lze předpokládat, že rozdíly v zastoupení obsahu mastných kyselin mezi systémy chovu (klec vs.

výběh) jsou spojeny s obsahem intramuskulárního tuku a příslušným obsahem fosfolipidů, které jsou typické pro jejich obsah n-6 a n-3 mastných kyselin. Co se týče například hustoty osazení ustájovacích prostor, je tímto profil mastných kyselin ovlivněn do určité míry. Při nižší hustotě osazení než 16 králíků/m^2 byl pozorován obecný pokles obsahu SFA a MUFA, zatímco byl zaznamenán nárůst PUFA (Dalle Zotte a kol., 2010). Chemické složení masa je definováno dalšími parametry, jako je například zastoupení tuku ve svalové tkáni. Existují úzké korelace mezi množstvím tuku v těle a obsahem tuku v mase a mezi obsahem vody a obsahem tuku v mase. Králíci chovaní ve velkých skupinách měli signifikantně nižší tukové krytí než ti chovaní ve skupinách malých. Stejně tak maso králíků chovaných ve velkých skupinách bylo chudší na tuk a obsahovalo více vody (Dal Bosco a kol., 2002; Combes a kol., 2010). Pro obsah bílkovin ani obsah popela nebyl zjištěn vliv velikosti skupiny (Szendrő a kol., 2009). Ve studii Krunta a kol. (2022) také nebyl zjištěn významný rozdíl pro chemické složení masa z hlediska obsahu tuku, bílkovin, vody nebo sušiny mezi boxovým a klecovým systémem ustájení při výkrmu králíků. Lišil se pouze obsah popelovin, které byly vyšší ve výběhovém systému než v klecovém.

Podmínkami ustájení jsou také ovlivněny textura masa a struktura svalových vláken. Co se týče složení svalu, Lefaucher (2010) uvádí, že svaly se skládají ze svalových vláken, typu I (βR) a typu II (αR, αW). U králíků jsou nejvíce zkoumány dva nejdominantnější svaly (*biceps femoris* a *longissimus thoracis et lumborum*). Svalová vlákna ovlivňují vývoj postmortálních změn, přičemž kvalita masa je také ovlivněna (Hernández a kol., 2006). Dvě základní charakteristiky, které definují svalová vlákna, jsou jejich průměr a obvod. Velikost svalu je ovlivněna také těmito dvěma charakteristikami, přičemž plocha průřezu je třetím rozhodujícím faktorem (Chodová a kol., 2014). Vliv hustoty osazení na charakteristiky svalových vláken v svalu *biceps femoris* byl hodnocen Volkem a kol. (2014). V této studii byl u králíků chovaných při hustotě obsazení 10 králíků/m^2 oproti 4 králíkům/m^2 podíl vláken typu αW 79,3 % oproti 59,2 %. Při srovnání stejných hustot osazení byl podíl vláken typu αR 24,5 % oproti 14,2 %. Stejně tak byla distribuce vláken typu βR vyšší při nižší hustotě osazení a činila 16,3 %, zatímco při vyšší hustotě osazení byla pouze 6,5 %. Tyto výsledky byly vysvětleny vyšší fyzickou aktivitou králíků, jak bylo popsáno i ve studii Krunt a kol. (2022). Plocha svalových vláken typu βR byla signifikantně menší u králíků chovaných při nižší hustotě osazení (1 882 oproti 2 744 μm^2), zatímco plocha vláken typu αR a αW zůstala téměř nezměněna v závislosti na hustotě osazení.

Co se týče senzorického zhodnocení králičího masa ve vztahu k systému ustájení, vědecká literatura je konzistentní ve vlivu ustájení, přes obsah tuku, na profil mastných kyselin, který je také ovlivněn prostřednictvím výživy (Volek a kol., 2018). Jednoznačně se v senzorických testech liší různá plemena králíků (Fadare a Arogbo, 2015) a jako signifikantní je považován vliv věku zvířat (Gondret a kol., 1998). U králíků starších než 84 dnů již dochází ke kombinaci efektů jako je chování, vliv prostředí, genetika, pohlaví a jiné z důvodu odlišného např. agresivního chování dle pohlaví, rozdílnému ukládání depotního tuku dle pohlaví atd. (Krunt a kol., 2022). Navíc u králíků dochází většinou k hodnocení masa vařeného, kde jsou rozdíly, například v barvě, minimální.

4.3 Vliv genotypu a systému ustájení na chování samic a kvalitu života jejich mláďat

Vliv genotypu samic v reprodukci na jejich chování nebyl ve vědecké literatuře prakticky zkoumán. Je obecně známo, že v intenzivních chovech jsou využívání králíci hybridních kombinací, kteří splňují nároky na produkci. Naopak v chovech na malých farmách jsou často využívání králíci původních plemen. Ve studii Krunta a kol. (2023) byl porovnán užitkový hybrid Hyplus s původním plemenem meklenburský strakáč, a to s výsledkem zjištění vyšší agresivity u samic Hyplus. Autoři studie se domnívají, že vyšší incidence agresivních událostí mezi samicemi Hyplus v porovnání s meklenburskými strakáči může být zdůvodněna selekcí na agresivní chování na malých farmách, kde se krmí většinou ručně a ošetřovatel má větší kontakt se zvířaty. Naopak, komerční hybridi jsou striktně selektováni pouze na produkční a reprodukční ukazatele, na rozdíl třeba od králíků v hobby chovech (Rooney a kol., 2014). Agresivní chování samic je problematické především pro jejich soužití ve skupinových ustájovacích systémech, a to nejen z pohledu sníženého welfare samotných samic a možných úhynů, či nutnosti vyřazení samice, v důsledku zranění, ale také kvůli zraňování nebo zabíjení mláďat cizích samic. To je běžné i u divokých králíků (Mykytowycz a Dudzinski, 1972), není to tedy nijak výjimečný stav v uměle vytvořeném prostředí, avšak zde s ekonomickými důsledky. Ve studii Krunta a kol. (2023) bylo pak pozorováno, že nejvyšší míra úhynu mláďat byla pozorována u samic Hyplus v porovnání s meklenburským strakáčem, a to v souvislosti se zraněními na těle. Značný počet mláďat uhynul již v hnízdě, a to v důsledku kompetitivních bojů v hnízdech, či při ochraně hnízda před cizí samicí. Autoři také zjistili, že pokud měly samice k dispozici nory (nepevný podklad k hloubení nor), nacházelo se v těchto systémech méně zraněných samic, případně mláďat v porovnání se systémy bez nor (na hluboké podestýlce). Zraněné samice se ale vyskytovaly v obou systémech ustájení a žádná studie nereportuje takový systém chovu, který by generoval nezraněné samice v rámci chovných

skupin. Například Gerencsér a kol. (2019) testovali několik modulů k chovu samic ve skupině a jejich slučování 18 dní po porodu, s výsledkem 50 % zraněných samic. Dále pak Rommers a de Greef (2018) vytvořili skupiny samic po 5 kusech a zjistili u nich 34 % zraněných kusů hned po hierarchickém formování skupiny a 53 % zraněných králíků po okocení.

Pokud jde o sledování krevních parametrů v rámci vlivu systému ustájení, Zita a kol. (2023) nepozorovali mezi zvolenými systémy (např. hluboká podestýlka, možnost hloubení nor, roštová podlaha, modifikace se zrcadly atd.) v rámci hematologických parametrů (hematokrit, hemoglobin, erytrocyty, leukocyty, neutrofily, monocyty, basofily a jiné) žádné rozdíly, přičemž výsledné hodnoty splňovaly referenční rozmezí hodnot pro daný druh. Autoři však zjistili signifikantní rozdíly pro jednotlivé biochemické parametry, jež ustájením ovlivněny byly. Hodnoceny byly například parametry jako glukóza, kreatinin, močovina, celkový protein, albuminy, globuliny, alkalin-fosfatáza a cholesterol. V této studii byl zjištěn vliv využití zrcadel v systémech ustájení samic, a to takový, že samice ustájené se zrcadly měly v krvi více kreatininu než samice v ustájeních bez zrcadel. Naopak, vliv využití zrcadel nebyl v této studii zaznamenán na obsah cholesterolu v krvi, ale Musco a kol. (2019) uvedli, že v jejich studii měli králíci, se zrcadly v ustájení, nejvyšší obsah cholesterolu v krvi. Obě studie ale nezaznamenaly žádné výkyvy v hodnotách, které by znamenaly nemoc nebo chronický stav u studovaných zvířat.

Jak bylo zmíněno již v kapitole výkrmu, systém ustájení výrazně modifikuje kvalitu kostí. Například Buijs a kol. (2014) jako jedni z mála autorských kolektivů hodnotili kvalitu kosti stehenní a lýtkové u samic v reprodukci v závislosti na systému ustájení. Došli k závěru, že samice z klecí měly vyšší lámavost kostí než samice z různých skupinových výběhových systémů. Také Zita a kol. (2023) zjistili, že pravděpodobně čím více fyzické aktivity spojené s používáním končetin králík má, tím je pevnost kosti vyšší. Proto nejvyšší hodnoty naměřili u kostí samic ustájených v různých formách výběhu a nejnižší pak v combi-parkovém systému, kde byla menší plocha pro pohyb. Combi-parkový systém je v současné době jedním z hojně užívaných systémů ustájení pro samice v reprodukci, pokud jde o alternativy ke klecovému systému chovu.

5 Závěr

Výsledky této doktorské disertační práce jsou logicky shrnuty v rámci výsledků, diskusí a závěrů jednotlivých publikací. V rámci této práce byli studováni jak králíci ve výkrmu, tak samice v reprodukci. Konkrétně pak z pohledu vlivu systému ustájení na produkční parametry, kvalitu masa, zdraví a welfare, případně u samic z pohledu vlivu ustájení a genotypu na chování, počty zranění a úhyny u mláďat. Dále pak byl zhodnocen jejich welfare a zdraví.

Lze konstatovat, že při volbě systému ustájení pro výkrm je třeba brát v potaz především konkrétní legislativu, možnosti trhu a profil cílového zákazníka. Na základě výsledků je možné doporučit skupinové systémy ustájení pro výkrm králíků. A to jednoznačně z pohledu welfare, avšak s ideální délkou výkrmu do 10 týdnů věku. Králíci ze skupinových systémů mají v porovnání s těmi z klecí sice nižší porážkovou hmotnost, ale vyšší podíl stehen, pevnější kosti a mají k dispozici více prostoru pro pohyb. Nutno však poznamenat, že existují studie, které reportují nevýznamné rozdíly pro porážkovou hmotnost mezi králíky z klecí a boxů. Nabízí se tedy spíš vysvětlení, že pokud rozdíly existují. Jedná se pravděpodobně o nezvládnutý management chovu (málo krmných míst, velká skupina zvířat atd.) Z hlediska kvality masa, existují rozdíly mezi jednotlivými systémy, avšak se nezdají být klíčové pro budoucí výzkum v rámci ustájovacích možností. Toto tvrzení lze podpořit i neprůkaznými rozdíly v senzorické analýze při hodnocení masa králíků z klecí a boxů. Je tedy zřejmé, že senzorickými vlastnostmi na základě ustájení nelze oslovit spotřebitele, což se o welfare prvcích chovu říct nedá. Na druhou stranu, čím vyšší welfare v chovech bude, tím vyšší bude cena finálního produktu. Je třeba si ale také uvědomit, že se blíží konec klecových chovů (viz situace kolem kožešinových zvířat a konec klecových chovů nosnic v ČR od roku 2027) a s tím i naléhavost výsledků této a jiných prací s ohledem na přizpůsobení se novým podmínkám.

Co se týče ustájení samic, agresivní chování nebude do budoucna možné zcela eliminovat. Vliv genotypu je sice cenným výsledkem, avšak pro intenzivní chovy je toto pravděpodobně neaplikovatelné. Jistý benefit z těchto výsledků mohou mít malé chovy, kde jsou původní plemena používána, případně další studie, které by si daly za cíl původní tezi rozvíjet dál. Na základě výsledků práce je zřejmé, že jsou-li samice v reprodukci hodně zaměstnané svým okolím, které nějakým způsobem simuluje jejich původní podmínky života, jsou agresivní méně, s čímž souvisí i agresivita projevovaná vůči mláďatům a tím pádem i jejich úhyny.

Z praktického hlediska se jeví jako nejlepší alternativou výkrmu králíků v klecích výkrm králíků ve skupině v tzv. combi-parcích. Combi-parky umožňují králíkům vyšší míru

naplnění druhově specifických aktivit, jsou relativně jednoduché na obsluhu a výkrm ve skupině je realizován v rámci vrhu sourozenců. V případě, že je dodržena délka výkrmu do cca 10 týdnů věku zvířat, nemělo by docházet k výskytům agonistických interakcí a zraněním.

V rámci přiložených článků byly buď potvrzeny nebo vyvráceny hypotézy jednotlivých experimentů a tím byly zároveň naplněny cíle práce.

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