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Caracterización nutricional de la carne ovina en sistema pastoril: Contenido de minerales, hierro hemínico, vitamina B12 y ácidos grasos del músculo *longissimus thoracis* en diferentes genotipos

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Doctorado en Ciencias Agrarias

Setiembre, 2024

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Resumen

La carne desempeña un papel importante en una dieta equilibrada y saludable para el humano debido a su riqueza nutricional. En este trabajo caracterizar la composición nutricional de la carne de cordero en un sistema de producción pastoril. Se determinó el contenido de macroelementos, Ca, Mg, Na y K, minerales traza como Se, Co, Zn, Cu, Mn, hierro total (TFe), hierro hemínico (HFe) y hierro no hemínico (NHFe), vitamina B12 y la composición en ácidos grasos de los glicerolípidos y glicerofosfolípidos del músculo *longissimus thoracis*, de diferentes biotipos genéticos producidos en Uruguay, merino Dohne (MD), Highlander® (H), Corriedale Pro (CPRO), Corriedale (C), crusa Merino Australiano x Corriedale (CxMA). El genotipo Merino Dohne tuvo la mayor cantidad de Ca ($66,6 \pm 6,3 \text{ mg.kg}^{-1}$), Highlander® y Merino Dohne la mayor cantidad de Mn ($304,1 \pm 26,0$ y $308,7 \pm 23,6 \text{ } \mu\text{g.kg}^{-1}$, respectivamente) que los demás genotipos. El contenido de HFe y la relación HFe/TFe fue mayor en los genotipos Corriedale y Corriedale Pro ($15,7 \pm 0,6$ y $15,4 \pm 0,7 \text{ mg.kg}^{-1}$ y $81,7 \pm 2,8 \%$ y $76,0 \pm 2,2 \%$, respectivamente) y, en consecuencia, menor NHFe, en relación con los otros grupos. También se obtuvo un mayor contenido de Zn en Corriedale ($32,6 \pm 1,3 \text{ mg.kg}^{-1}$). Los resultados no mostraron diferencias significativas entre genotipos en la composición en ácidos grasos de la carne, excepto para los ácidos grasos relevantes como C16:0 (MD, C), C18:3n3 (H, C) y CLA (H, CPRO, CxAM) para glicerolípidos. También C18:1 (H, CPRO y CxAM), C18:2n6 (H, CxAM) y C18:3n3 (H, C) para glicerofosfolípidos. Asimismo, se destaca el mayor contenido de ácidos grasos anteiso (en RM y MD) y la mejor relación del índice h/H para MD y C. Para las actividades enzimáticas del metabolismo de los ácidos grasos, el genotipo MD mostró una menor actividad de la enzima desaturasa Δ-9 para C16:0 que C, CPRO y CxAM. Además, H mostró una menor actividad de la enzima Δ-6 desaturasa que C y tanto MD como CxAM mostraron una menor actividad de la enzima elongasa que C. Se obtuvieron valores importantes en el aporte de algunos minerales y ácidos grasos con respecto para una dieta equilibrada y deja un margen para seguir investigando.

Palabras claves: genética, carne de cordero, minerales, ácidos grasos, pastura

Nutritional characterisation of sheep meat in pastoral system: Mineral, heme iron, vitamin B12 and fatty acid content of *longissimus thoracis* muscle of different genotypes

Summary

Due to its nutritional value, meat plays an important role in a balanced and healthy human diet. For this reason, this work proposed to study the nutritional composition of lamb meat in a pastoral production system. The content of macroelements, Ca, Mg, Na and K, trace minerals such as Se, Co, Zn, Cu, Mn, total iron (TFe), heminic iron (HFe) and non-heminic iron (NHFe), vitamin B12 and the fatty acid composition of glycerolipids and glycerophospholipids of the *longissimus thoracis* muscle were determined, of different genetic biotypes produced in Uruguay, Merino Dohne (MD), Highlander® (H), Corriedale Pro (CPRO), Corriedale (C), Australian Merino x Corriedale cross (CxMA). The Merino Dohne genotype had the highest Ca (66.6 ± 6.3 mg.kg⁻¹), Highlander® and Merino Dohne the highest Mn (304.1 ± 26.0 and 308.7 ± 23.6 µg.kg⁻¹, respectively) than the other genotypes. HFe content and HFe/TFe ratio were higher in the Corriedale and Corriedale Pro genotypes (15.7 ± 0.6 and 15.4 ± 0.7 mg.kg⁻¹ and 81.7 ± 2.8 % and 76.0 ± 2.2 %, respectively) and correspondingly lower NHFe, relative to the other groups. A higher Zn content was also obtained in Corriedale (32.6 ± 1.3 mg.kg⁻¹). The results showed no significant differences between genotypes in the fatty acid composition of the meat, except for relevant fatty acids such as C16:0 (MD, C), C18:3n3 (H, C) and CLA (H, CPRO, CxAM) for glycerolipids. Also, C18:1 (H, CPRO and CxAM), C18:2n6 (H, CxAM) and C18:3n3 (H, C) for glycerophospholipids. Also, the higher content of anteiso fatty acids (in MR and MD) and the better h/H ratio for MD and C are highlighted. For fatty acid metabolism enzyme activities, the MD genotype showed lower Δ-9 desaturase enzyme activity for C16:0 than C, CPRO and CxAM. In addition, H showed lower Δ-6 desaturase enzyme activity than C, and both MD and CxAM showed lower elongase enzyme activity than C. Important values were obtained in the contribution of some minerals and fatty acids with respect to a balanced diet and leaves a margin for further research.

Keywords: genetic, lamb meat, minerals, fatty acids, pasture

1. Introducción

1.1. Marco teórico

Uruguay forma parte de la principal región agroexportadora de alimentos del mundo, junto con Argentina, Brasil y Paraguay. Es un país esencialmente agropecuario: el 93 % de su superficie es apta para la producción agrícola ganadera, en donde la actividad agropecuaria ocupa el 80% de aquella (Uruguay XXI, 2021). Con un mercado interno reducido, la creciente producción del sector agroindustrial del país se destina a la exportación. En 2019, el sector agroindustrial constituía el 82 % del total de los bienes exportados. En ese año, las ventas agroindustriales, considerando solo el sector alimenticio, ascendieron a US\$ 5.132 millones, equivalente al 68 % de lo exportado. Carne bovina, lácteos, soja y arroz fueron los principales alimentos exportados por Uruguay (Uruguay XXI, 2021).

Nuestro país, se posiciona como uno de los diez principales exportadores de carne ovina en el mundo y el primero en la región (Trade Map, 2022). La producción de carne ha pasado por diferentes etapas en los últimos años, con un stock a la fecha de 6,230 millones de cabezas, con una producción de 73.000 toneladas de carne (Ministerio de Ganadería, Agricultura y Pesca-Dirección de Estadísticas Agropecuaria [MGAP-DIEA], 2021). El ingreso por carne ovina fue de 98,5 millones de dólares, representando un 3 % de los ingresos totales del sector (Instituto Nacional de la Carne [INAC], 2019).

La producción ovina se concentra en la unidad geomorfológica cuesta basáltica en predios ganaderos extensivos donde la base nutricional es campo natural sobre suelos superficiales de basalto, siendo el producto principal la lana fina, y el secundario, la carne de cordero (Baeza et al., 2019). En sistemas intensivos y semiintensivos, como, por ejemplo, aquellos que se asocian a la rotación cultivo-pastura, aprovechando el buen potencial del forraje que en estas condiciones se produce, la venta de corderos con sus diferentes alternativas (cordero precoz, cordero pesado y superpesado) constituyen el principal ingreso. Estos sistemas actualmente corresponden a zonas del país donde el rubro interactúa con sistemas agrícolas ganaderos, lecheros, hortícolas y frutícolas, donde se han desarrollado genotipos

carniceros y genotipos doble propósito. Es así que, en los últimos veinte años, si bien el stock ovino ha disminuido, ha aumentado el número de productores en el sur, como resultado de la reconversión de predios hortícolas y frutícolas, vitivinícolas, o en predios lecheros como un rubro adicional (Ganzábal et al., 2003), lo cual ha tenido un importante impacto social y económico en el medio rural del país.

Como consecuencia de la variabilidad de la demanda del mercado externo y los precios competitivos, el país ha puesto énfasis en aumentar el valor agregado de los productos, para ser más competitivo en los mercados internacionales.

Es así que Uruguay asumió su compromiso de preservar un sistema productivo sustentable, a cielo abierto. Esto, sumado al empleo responsable de la tecnología productiva, se asocia a una imagen de bienestar animal, producto natural, seguro y altamente nutritivo, cuya explotación es importante como promoción de la exportación (Instituto Nacional de la Carne [INAC], 2022).

Según Burlingame et al. (2012), el 70 % de la población mundial en 2050 será urbana. Basados en los cambios en la sociedad, como resultado de la migración rural y, en algunos casos, del aumento de ingresos per cápita, conducirían al crecimiento sustancial del consumo de carne como fuente estratégica de proteína en la dieta humana. El crecimiento esperado en la producción y consumo de carne ovina será impulsado en su mayoría por los países en desarrollo. Esto determinará un aumento de la demanda, hacia países asiáticos, por ejemplo, siendo uno de los principales destinos, el MENA (norte y medio este de África). Otros potenciales importantes importadores serían los países árabes para los cuales la carne ovina es parte de la cultura. Con el desafío de nuevos mercados y consumidores con exigencias particulares, toma importancia la necesidad de valorizar la carne ovina uruguaya tanto de biotipos laneros como de doble propósito aumentando el valor del producto y mejorando su competitividad, lo cual podría permitir el acceso a otros mercados más atractivos económicamente. El conocimiento de los atributos de la carne de cordero producido por diferentes biotipos genéticos, en condiciones de pastura en diferente zona del país, es necesario para caracterizar estos sistemas, valorizar y diferenciar el producto carne de cordero desde un enfoque de calidad, novedoso y de gran impacto en el rubro ovino.

2. Antecedentes

2.1. La carne como alimento

La ingestión de alimentos frescos sanos y saludables juega un papel crucial en el mantenimiento de la salud humana. El término *dieta equilibrada* ha ganado una inmensa popularidad en todo el mundo, debido a la creciente concientización sobre el mantenimiento del estado de salud entre las poblaciones (Ahmad et al., 2018). Una dieta equilibrada garantiza la ingesta de todos los nutrientes esenciales que el cuerpo humano necesita para realizar las funciones de la vida diaria (Eze et al., 2017; Organización de las Naciones Unidas para la Agricultura y la Alimentación-Organización Mundial de la Salud [FAO-OMS], 1998). En este contexto, el conocimiento de la composición nutricional de los alimentos se ha convertido en un factor importante a la hora de ingerir una comida equilibrada, lo que, a su vez, contribuye a un buen estado de salud de las personas. Todos los nutrientes requeridos por el humano son suministrados por una serie de alimentos como la carne, los cereales, la leche, las frutas y las verduras. Entre ellos, las carnes en general ocupan un lugar destacado, ya que satisfacen la mayor parte de las necesidades proteínicas de los seres humanos (Ahmad et al., 2018), particularmente en los individuos con altas necesidades como niños, ancianos y mujeres gestantes (Ministerio de Salud Pública [MSP], 2021; Moraes et al., 2021).

Si bien existen estudios epidemiológicos sobre el consumo de carnes rojas, en donde se ha señalado una posible relación entre su consumo y los elevados riesgos de padecer enfermedades cardiovasculares, diversas formas de cáncer y trastornos metabólicos, no se puede ignorar su papel en la evolución de la especie humana, concretamente en su desarrollo cerebral e intelectual (Hawkes, 2001; Pereira y Vicente, 2013; Puerta et al., 1992). La carne contiene aminoácidos de alto valor biológico, micronutrientes como hierro (Fe), principalmente Fe hemo, selenio y zinc, y vitaminas del complejo B —entre ellas, alto contenido de B12—, importantes para el desarrollo del sistema nervioso (Geay et al., 2001; Ortigues-Marty et al., 2005), ácidos grasos como los ácidos grasos insaturados (AGPI) y, dentro de estos, los ácidos grasos de la familia de los ω-3, y compuestos bioactivos que hacen de la carne un

alimento de suma importancia en la dieta de los humanos (Belhaj et al., 2021; Bohrer, 2017; Cabrera et al., 2015; Cabrera y Saadoun, 2014; Purchas et al., 2004; Purchas et al., 2014; Schönfeldt et al., 2013).

2.2. Principales nutrientes de la carne

2.2.1. Proteínas y aminoácidos

La carne es fuente de proteína por excelencia junto a otros productos de origen animal como huevo, leche y pescado. La proteína de la carne provee aminoácidos necesarios para construir y mantener los tejidos del cuerpo, así como para contribuir en los procesos de regulación metabólica (Charrondière et al., 2013; Jacob y Pethick, 2014; Williams, 2007). Se destaca especialmente por un mayor contenido en aminoácidos esenciales (AEE) y, sobre todo, en aminoácidos de cadena ramificada (BCAA), valina, isoleucina y leucina. Las diversas fuentes de proteínas de origen animal de la dieta humana se caracterizan por su alto valor biológico, en el que influyen el contenido de EAA, su digestibilidad y facilidad de absorción (Gutierrez, 2000; Pereira y Vicente, 2013). El valor biológico de la carne está en el rango 75-85 % y la digestibilidad de la proteína está entre 92-100 %, aportando la carne magra y cruda entre 19-23 % de proteína total (Ahamed et al., 2018; Pereira y Vicente, 2013; Warriss, 2010). El valor nutritivo de la carne puede variar de forma importante en función de la presencia o ausencia de determinados aminoácidos (Ponnampalam et al., 2019; Santé-Lhoutellier y Pospiech, 2016).

Estudios recientes han revelado que la principal diferencia en la proporción de aminoácidos esenciales está dada por la genética, la edad del animal y tipo de músculo. Gutierrez (2000) y Ke et al. (2023) reportaron que los contenidos de, por ejemplo, valina, isoleucina, fenilalanina, arginina y metionina en el músculo aumentaron con la edad del animal. La mayor proporción de proteínas en el músculo la constituyen las miofibrillas, seguidas por las proteínas sarcoplásmicas que forma parte de las enzimas musculares y la mioglobina, siendo las primeras las de mayor valor nutritivo (Gutierrez, 2000; Jacob y Pethick, 2014). El contenido de aminoácidos esenciales difiere según las distintas partes de la canal, asociado al predominio de 0determinadas

fibras musculares. Es así que la carnosina (beta-alanil-L-histidina), poderoso escudo protector frente a los radicales libres, se encuentra en mayores concentraciones en las fibras musculares blancas del tipo IIb (Aristoy y Toldrá, 1998; Cornet y Bousset, 1999), involucradas en el metabolismo energético en músculos de contracción rápida (Abe, 2000).

2.2.2. Lípidos y ácidos grasos

Los lípidos son fuente de energía y ácidos grasos esenciales, además de contribuir con el transporte de las vitaminas liposolubles (Aberle et al., 2012). La cantidad de lípidos en la carne está distribuida por todo el músculo —grasa intramuscular, intermuscular, cavitaria y subcutánea— (Williams, 2007).

La grasa intramuscular hace referencia a la grasa contenida entre las fibras musculares, contribuye en el *marmoreo*, el cual está relacionada con las características de la carne como sabor, terneza y jugosidad, siendo estas las mayores características de aceptabilidad de la carne por parte del consumidor (Realini, Pavan, Johnson et al., 2021; Realini, Pavan, Purchas et al., 2021), así como su composición de ácidos grasos por estar relacionada con la salud humana (European Food Information Council [EUFIC], 2015; Higgs, 2000).

La grasa intramuscular es uno de los componentes más variables, reportándose valores en cordero entre 0,91 % a 9,5 % (Craigie et al., 2017; Pannier, Pethick, Geesink et al., 2014). Esta gran variación se debe a factores como genética, tipo de músculo, alimentación, sistema de producción (Belhai et al., 2020; Nuernberg et al., 2008; Santé-Lhoutellier y Pospiech, 2016; Williams, 2007).

Los ácidos grasos presentes en la carne de rumiantes son consecuencia de la dieta ingerida, mayoritariamente ácidos grasos insaturados (dobles enlaces carbono-carbono) y del proceso de biohidrogenación de los ácidos grasos por los microrganismos del rumen (Buccioni et al., 2012; Elizalde et al., 2020). Los dobles enlaces en los ácidos grasos insaturados se presentan en la configuración cis. En rumiantes, como resultado de la biohidrogenación en el rumen, existe una importante cantidad de dobles enlaces en configuración trans. La posición de la insaturación (doble enlace) se indica con la letra griega omega (ω), también se acepta el uso de la

letra ene (n) y un número que designa en que enlace se encuentra la insaturación contando a partir del carbono metilo (Mc Donald et al, 2011). El mayor ácido graso con configuración trans es el ácido vaccénico 18:1 n-7, que es un producto intermediario de la biohidrogenación del 18:2 n-6 (linoleico), el cual, a su vez, es convertido en el ácido linoleico conjugado (ALC, 18:2 cis-9, trans-11).

Dentro de los ácidos grasos esenciales, el primero en importancia es el ácido linoleico (18:2 n-6) perteneciente a los AGPI. Es derivado enteramente de la dieta (granos, cereales y oleaginosos) y, en transformado monoinsaturados y saturados por el proceso de biohidrogenación llevado adelante por la microflora ruminal, solo un 10 % pasa sin modificar para ser incorporado en los tejidos. Segundo en importancia sigue el ácido alfa-linolenico 18:3 n-3 (ALA) (AGPI), el cual es el principal ácido graso que aparece en las dietas forrajeras y sus subproductos (McDonald et al., 2011; Wood et al., 2008), si bien no en los tejidos, ya que, según trabajos reportados por Doreau y Ferlay (1994), el ácido linolénico es biohidrogenado entre un 85 % y un 100 %, mientras que el ácido linoleico lo es en un 70-95 %, por lo que queda más sin ser biohidrogenado para ser incorporado a los tejidos.

Es así que los sistemas de terminación con dietas forrajeras producen mayores cantidades de AGPI n-3 en el músculo que la alimentación en base a concentrados, lo que puede mejorar tanto las proporciones AGPI:AGS como de n-6:n-3 (Gatellier et al., 2005; Pordomingo et al., 2012; Rochfort et al., 2008; Saadoun et al., 2013). Los sistemas de alimentación en base a pasturas aumentaron el porcentaje de ácidos grasos de la familia n-3 en los lípidos del músculo *longissimus* tanto de la carne de vacuno como de cordero (Demirel et al., 2006; Scollan et al., 2006). Contrariamente, en corderos consumiendo concentrados aumentó de forma significativa la relación n-6:n-3, en comparación con los animales alimentados con forraje (Ponnampalam et al., 2002).

La carne proveniente del ovino y de otros rumiantes históricamente ha sido asociada con el incremento del riesgo de desarrollar enfermedades cardiovasculares (ECV), debido a su alta proporción de ácidos grasos saturados (AGS), particularmente ácido mirístico (14:0) y ácido palmítico (16:0) que tienen propiedades de aumentar las lipoproteínas de baja densidad (Al-Shaar et al., 2020; Chikwanha et al., 2018; Prache

et al., 2022; Salter, 2013). Sin embargo, nuevas evidencias muestran que la carne magra (no procesada) de rumiantes, como la del ovino, es una fuente importante de ácidos grasos fisiológicamente funcionales y potencialmente beneficiosos para la salud (Cabrera y Saadoum, 2014; Nuerenberg et al., 2008). Ejemplo de esto son los ácidos grasos poliinsaturados (AGPI) de la familia de los ácidos grasos omega-3 (n-3) y algunos intermediarios de la biohidrogenación de algunos AGPI, como el ácido ruménico (cis-9, trans-11 C18:2) y ácido transvaccénico (t-11 C18:1), que muestran efectos benéficos para la salud humana (Chikwanha et al., 2018; Realini, Pavan, Purchas et al., 2021; Vahmani et al., 2020). Los efectos atribuidos al ALC son múltiples, entre ellos cabe destacar su papel como agente antitumoral (Bruen et al., 2017; Kelley et al., 2007; Shokryazdan et al., 2017), y antiarterioesclerótico (Dilzer y Park., 2012; Kim et al., 2016).

2.2.3. Carbohidratos

La cantidad de carbohidratos en los mamíferos representa tan solo el 1-2 % de la masa muscular (Bello, 2010). La principal fuente de carbohidrato en el animal es el glucógeno (polímero de alfa-D-glucosa), el cual se almacena principalmente en el hígado y los músculos, pero también en glándulas y órganos en menor medida, así como cantidades importantes en la sangre de glucosa (Ahmad et al., 2018; Bello, 2010). Luego de la faena, se da el proceso, de transformación de músculo a carne en donde se producen cambios importantes como consecuencia de la actividad metabólica en el tejido; al final de ese proceso se tiene como producto la formación de ácido láctico y cantidades vestigiales de metabolitos con estructuras de azúcares como glucosa, glucosa-6-fosfato, entre otros azúcares-fosfatos; todas estas modificaciones se rigen por hormonas y enzimas (Bello, 2010; Jensen et al., 2011). A pesar de que la carne presenta una concentración de carbohidratos muy baja para el consumo humano, estos son muy importantes, ya que pueden afectar drásticamente las características metabólicas *perimortem* sobre todo a lo referido la tasa de descenso y valor final de pH, que influyen de forma indirecta en las características como el color, la textura, la terneza y la capacidad de retención de agua (Jensen et al., 2011). La cantidad de glucógeno muscular se ve afectada por la especie y el tipo de fibra muscular, entre

otros, y es por ello que se ha prestado mucha atención a comprender y controlar los niveles adecuados de glucógeno prefaena (Pösö y Poulanne, 2005).

2.2.4. Minerales

Los minerales juegan un rol vital en varios procesos biológicos en los animales que influyen en su desempeño, reproducción, inmunidad y supervivencia. Estos procesos pueden ser alterados cuando los minerales están fuera de los rangos adecuados (Suttle, 2022), estando, además, ligados directa o indirectamente a la salud de los consumidores. Los minerales son esenciales o tóxicos, dependiendo de las funciones y cantidad en que se encuentran en la matriz del alimento (Schmitt et al., 2014). Estos se pueden categorizar, en función de los requerimientos, en macrominerales, que incluyen sodio (Na), calcio (Ca), fósforo (P), cloro (Cl), potasio (K) y azufre (S), y minerales traza, que se refieren a los requeridos en menor cantidad, y estos incluyen hierro (Fe), zinc (Zn), iodo (I), cobre (Cu), cobalto (Co), manganeso (Mn), selenio (Se) y flúor (F) (Belhaj et al., 2021; Cabrera y Saadoun, 2014; Kasap et al., 2018; Ramos et al., 2012; Soetan et al., 2010).

Según Henderson et al. (2003), el Ca y el Mg no están presentes en cantidades altas en la carne, pero sí lo están el K y el P. Sin embargo, la carne sí es fuente importante de minerales traza esenciales, como el Fe, Se, Zn, Cu, Mn y Co, los cuales son cofactores del sistema enzimático antioxidante y, sobre todo, claves en las metaloenzimas y su actividad para contrarrestar los radicales libres en el organismo (Wang y Fu, 2012).

El Fe es requerido para la síntesis de hemoglobina, mioglobina y algunas enzimas (Cabrera et al., 2010), tanto para los animales como para el humano. Existen dos formas principales de Fe encontrada en los alimentos, el Fe hemínico (HFe), derivado principalmente de la hemoglobina y mioglobina en el tejido animal, y el no hemínico (NHFe), que proviene principalmente de alimentos de origen vegetal, difiriendo entre ambos su biodisponibilidad (Pretorius et al., 2016; Zimmermann y Hurrell, 2007). Según Hallberg et al. (1997), en promedio, la absorción del HFe proveniente de la carne es de alrededor de un 25 % (10 %-40 %). La genética, tipo de

músculo, edad a la faena y fisiológica, son factores que influyen en el contenido de hierro hemínico en la carne (Pretorius et al., 2016).

El Zn, al igual que el Fe, se encuentra en cantidades elevadas en alimentos de origen animal, principalmente en la carne, que aporta el 40 % de las cantidades diarios requeridas por el hombre (Szefer y Grembecka, 2006). Cuando se sustituye la fuente de carne por vegetales, con estos últimos se están ingiriendo fitatos con los cuales el Zn forma enlaces químicos y, como consecuencia, se reduce la disponibilidad en el intestino al igual que el Fe (Pretorius et al., 2016; Szefer y Grembecka, 2006). En algunas regiones de África y Asia, el consumo de Zn es bajo, debido al tipo de dieta, con bajo consumo de carne en relación con el consumo de cereales, y la población presentar deficiencias importantes, sobre todo en niños (Osendarp et al., 2001; Reilly, 2008; Szefer y Grembecka, 2006).

La cantidad de Se en alimentos de origen animal son determinados por las cantidades encontradas en las plantas consumidas y el suelo donde se desarrollan (Szefer y Grembecka, 2006, Kabata-Pendias y Szteke, 2015). Las funciones nutricionalmente esenciales del Se son desempeñadas por unas veinticinco selenoproteínas. Las selenoproteínas específicas incluyen el glutatióperoxidasa, la tiorredoxina reductasa, las 5-yodotironina deiodinasa, la selenoproteína P y otras. La mayoría de los compuestos de Se orgánico e inorgánico solubles en agua presentes en los alimentos se absorben con relativa eficacia a través del tracto gastrointestinal (80 %-95 %), siendo las formas orgánicas (por ejemplo, los selenoaminoácidos, como la selenometionina y la selenocisteína) las que se absorben más fácilmente que las formas inorgánicas (Kabata-Pendias y Szteke, 2015). El Se está mayoritariamente unido a proteínas en los tejidos vegetales y animales; es por eso que las fuentes alimentarias más importantes de Se son las carnes, por su alto contenido en proteínas, y los cereales (por ejemplo, trigo grano duro), por su gran consumo (Jenny-Burri et al., 2010, Rose et al., 2010).

El Mn es un mineral esencial en la dieta de mamíferos. Tiene dos funciones biológicas: actúa como constituyente de metaloenzimas (arginasa, superóxido dismutasa, piruvato carboxilasa) y como activador de enzimas (metabolismo de aminoácidos, lípidos y carbohidratos) (European Food Safety Authority [EFSA],

2013). El aumento de las concentraciones de Mn inhibe la función metabólica de la enzima dependiente del Fe, por lo que largas exposiciones a exceso de Mn resultan en una deficiencia de Fe (anemia) (Kabata-Pendiasy Szteke, 2015). Los alimentos de origen animal presentan valores bajos de Mn, en un rango de 0,003 a 0,14 mg/100 g de carne, y, a pesar de la variación geográfica, son muy bajas las diferencias entre alimentos en diferentes regiones (Szefo y Grembecka, 2006).

Si bien la carne no es una fuente importante de Co debido a los bajos valores que esta contiene (<0,013 mg/100 g), es considerado un minera traz esencial para la salud de los mamíferos y especialmente importante para los bovinos y ovinos, ya que como rumiantes pueden sintetizar vitamina B12 y el Co forma parte de la misma (Szefer and Grembecka, 2006).

Estudios realizados por Patel et al. (2020) identificaron que el ambiente productivo y el régimen de alimentación como las variables de más impacto en el perfil de minerales en ganado. Prácticas de manejo (sistemas de pastoreo), así como el ambiente (déficit de agua, por ejemplo), pueden provocar que el animal consuma tierra, lo que le aporta una importante cantidad, por ejemplo, de Co, Fe, I, Se, entre otros (Whitehead, 2000).

El efecto de dietas forrajeras sobre el consumo de minerales y la concentración de estos en la carne es un importante espacio que aún necesita ser investigado en profundidad (Holman et al., 2021).

2.2.5. Vitaminas

Tanto la carne de bovinos como de ovinos es una valiosa fuente de vitaminas del complejo B (vitaminas solubles en agua), ya que aporta aproximadamente el 25 % de las cantidades dietéticas recomendadas por 100 g de carne de riboflavina, B3 (niacina) y vitamina B6 (pyridoxine) y casi dos tercios de las necesidades diarias de vitamina B12 (cobalamina) (Aberle et al, 2012; Williams, 2007; Wood, 2023). Las vitaminas de este grupo son necesarias como cofactores en muchas reacciones que intervienen en la producción de energía y en la síntesis de aminoácidos y ácidos grasos. En particular, la vitamina B12 es una vitamina de estructura compleja con un átomo central de cobalto unido a seis ligandos. Es sintetizada por bacterias que habitan en el

tracto gastrointestinal y es necesaria en el organismo como coenzima en el metabolismo del propionato y en la conversión de homocisteína en metionina.

Las vitaminas liposolubles, A, D, E y K, no están presentes en grandes cantidades en músculo, pero las cantidades de vitamina A son altas en el hígado (Henderson et al., 2003). Según este último autor, los hombres y las mujeres del Reino Unido obtienen el 34 % y el 22 %, respectivamente, de la vitamina A de la carne y los productos cárnicos en forma de retinol, que es necesario para el mantenimiento del crecimiento de la piel, la visión y el sistema inmune (Green y Fascetti, 2016; Juárez et al., 2021). Blanco et al. (2019) midieron vitamina A en músculo *longissimus thoracis* en corderos en dos sistemas de producción (en encierro y en pastoreo) durante el período de lactancia y obtuvieron diferencias significativas en luteína, retinol, alfa y gamma tocoferol, con mayores concentraciones en el músculo en los corderos provenientes de sistemas de producción en pastoreo. La vitamina D interviene en la homeostasis del Ca, formando parte de huesos y dientes, siendo la carne, una fuente de aporte medio (15 %) (Wood, 2023). La vitamina E es un antioxidante importante en el organismo, que protege contra los radicales libres producidos en las reacciones de oxidación durante el crecimiento y especialmente en el período *post mortem*, cuando la oxidación de los ácidos grasos puede producir cambios en el color del músculo (Bellés et al., 2019; Wood et al., 2004), en el sabor de la carne (Harris et al., 2001; Rowe et al., 2004) y la pérdida de agua muscular y, en consecuencia, afectar las características organolépticas de la carne y su vida útil (Dunshea et al., 2005). La vitamina K es coenzima de una carboxilasa, enzima requerida para la síntesis de proteína implicada en la coagulación de la sangre y metabolismo del hueso. Los microorganismos del rumen sintetizan grandes cantidades de vitamina K y solo se observa una deficiencia en presencia de un antagonista metabólico (AminiPour et al., 2011).

El valor nutricional de la carne, por tanto, puede ser modificado o influido por diferentes factores, como características de los alimentos (El-aal y Suliman, 2008; Fernandes Júnior et al., 2013), especie animal, biotipos (Hopkins, 2016), tipo de músculo (Ponnampalam et al., 2015), edad, sexo (Jacob y Pethick, 2014) sistema de

producción (Ates et al., 2020; Juárez et al., 2021; Pereira y Vicente, 2013; Santos-Silva et al., 2002) y manejo prefaena (Grandin, 2016) y posfaena (Przybylski et al., 2016).

Estas diferencias podrían permitir asociar, por ejemplo, un tipo de genética (razas o biotipos) con una composición nutricional determinada. La carne ovina, y de cordero en particular, proveniente de nuestros sistemas de producción a cielo abierto, ofrece una oportunidad de diferenciación y valorización desde un punto de vista de la composición nutricional, lo cual permite definir estrategias, con vistas al desarrollo de productos exportables diferenciados.

2.3. Visión del consumidor de los productos de origen animal

La carne es una fuente concentrada de nutrientes, considerada esencial para un óptimo crecimiento y desarrollo del humano (Higgs, 2000). De hecho, el consumo de carne —particularmente, roja— ha contribuido al desarrollo del tracto gastrointestinal humano, así como de los rasgos cráneo-dentales, del cerebro y de la postura bípeda, lo que permite diferenciar al hombre de otros homínidos (Larsen, 2003; Mann, 2007; Pereira y Vicente, 2013). A pesar de su riqueza nutricional, se le ha dado una imagen negativa al consumo de carne roja como promotora de enfermedades no trasmisibles, como el cáncer, enfermedades cardiovasculares (ECV), diabetes tipo II, entre otras (McDaniel et al., 2010; Orellana et al., 2009; Pereira y Vicente, 2023; Savoini et al., 2016), desestimulando su consumo (Krauss et al., 2000; Wood et al., 2004). No obstante en el año 2023 la FAO publicó un libro en defensa de los alimentos de origen animal, incluida la carne. En dicha publicación destaca la relevancia de la carne en una dieta saludable (Food and Agriculture Organization [FAO], 2023). En Uruguay hay una cultura de consumo de carne roja de bovina y ovina. El consumo de carne ovina venía con tendencia a la baja desde 2015 pero, en este último año 2023 aumentó a un consumo de 3,0 kg/habitante/año (INAC, 2024). En una encuesta realizada por Realini et al. (2022), de la población entrevistada el 85,7 % declaró comer carne bovina, mientras que solo un 4,6 % declaró comer carne ovina. (<2 kg/persona/año). Esta misma encuesta arrojó como resultado que la disminución del consumo estaba relacionada con el precio de la carne (46,1 %), por motivo de salud (21,4 %), y cambio en la dieta (19,4 %). Con respecto a la salud, hay visiones contrapuestas, pues algunos

consumidores reportan que no dejan de consumir carne por su valor nutricional —por la proteína— y otros declararon que la asociaban a una dieta no es saludable por aspectos como el ácido úrico, cáncer y colesterol. En ninguno de los casos se hace referencia a otros nutrientes de suma importancia para la salud humana como son el HFe y vitamina B12, entre otros (Realinni et al., 2022). Según estos autores, esto podría deberse a la falta de información unificada de las características nutricionales de las carnes roja y, en particular, la de cordero, producidas en las diferentes áreas agroecológicas del país. Sumado a esto, hay una creciente preocupación sobre la sostenibilidad de la producción en los nuevos escenarios más intensivos y sus posibles daños sobre el medioambiente y en aspecto de bienestar animal, que también hay que tener presente. Por todo esto, es importante profundizar en la investigación para que el producto carne en general y la de cordero en particular sean competitivos en los mercados cada vez más exigentes y, así, más consumidores puedan incorporar la carne de cordero a sus dietas. Con la idea de contribuir a caracterizar nutricionalmente la carne de cordero en el sistema tradicional de producción del país, esta tesis se enfocó en la composición de nutrientes claves para la vida humana, como los minerales, vitaminas y ácidos grasos de la carne proveniente de diferentes biotipos. Los resultados de estas investigaciones tendrán un impacto en el valor nutricional de un alimento único, como la carne, y así contribuir con la producción de calidad, la salud del consumidor y la valorización del ovino y del sistema productivo.

3. Hipótesis y objetivos

3.1. Hipótesis planteada

La carne de cordero producida en sistema pastoril presenta una composición nutricional de interés para la salud humana, por contener minerales, vitaminas y ácidos grasos, y podría variar según el genotipo.

3.2. Objetivos

3.2.1. General

Estudiar el estatus de macrominerales, elementos traza, formas de hierro y de la composición lipídica de la carne de corderos de diferentes genotipos provenientes de sistemas pastoriles en el litoral noroeste del Uruguay.

3.2.2. Específicos

- [1] Determinar el contenido de minerales Ca, K, Mg, Na, Co, Zn, Cu, Mn y Se en el músculo *longissimus thoracis* de corderos alimentados a pasturas.
- [2] Determinar las formas de hierro: hierro total (FeT), hierro hemínico y no hemínico (NHFe).
- [3] Determinar la composición en ácidos grasos de los glicerolípidos y glicerofosfolípidos del músculo *longissimus thoracis* de corderos alimentados a pasturas.
- [4] Estimar los índices de salud lipídica para los consumidores e índices del metabolismo de los ácidos grasos relacionados con las enzimas desaturadas, elongasas y tioesterasas.

3.3. Estrategia experimental

Para responder al objetivo general y a los objetivos específicos, durante la investigación se realizaron los siguientes estudios:

I. Se determinó el estatus de los minerales traza FeT, Zn, Cu, Mn, Co y Se y macrominerales Ca, Mg, Na y K, en el músculo *longissimus thoracis* de corderos Corriedale (C), Merino Dohne (MD), Corriedale pro (CPro), Highlander® (H) y crusa Merino Australiano x Corriedale (MAXC) alimentados a base de pasturas.

II. Se determinaron las formas de hierro total (FeT), hierro hemínico (HFe) y no hemínico (NHFe) en el músculo *longissimus dorsi* de corderos Corriedale, Merino Dohne, Corriedale Pro, Highlander® y crusa Merino Australiano x Corriedale en una dieta a base de pasturas.

III. Se determinó la composición en ácidos grasos de los glicerolípidos y glicerofosfolípidos y la actividad enzimática en el músculo *longissimus thoracis* de los corderos corriedale, merino Dohne, corriedale pro, Highlander®, Romney Marsh y crusa Merino Australiano x Corriedale en una dieta a base de pasturas.

IV. Finalmente se estimaron los índices lipídicos y de actividad enzimática a partir del perfil de ácidos grasos detectados, de importancia para la salud humana, en el músculo *longissimus thoracis* de los corderos Corriedale, merino Dohne, Romney Marsh, Corriedale pro, Highlander® y crusa Merino Australiano x Corriedale, alimentados a pasturas.

Este trabajo se divide en dos capítulos. En el primer capítulo se presentan los estudios realizados I y II y en el segundo, se presentan los estudios III y IV. Luego, una discusión general y conclusiones que se referirán al conjunto de los resultados obtenidos a lo largo de esta tesis.

Macrominerales, minerales traza y estado del hierro hem y no hem en
músculo *longissimus dorsi*, de cinco razas de corderos doble propósito criados en
sistema de pastoreo en Uruguay

Resumen

La producción de carne ovina se enfrenta a nuevos desafíos, por lo que el conocimiento profundo de los atributos de la carne de cordero producido por diferentes genotipos y en condiciones de pastura, son necesario para caracterizar estos sistemas, valorizar y diferenciar el producto desde un enfoque de calidad y hacia una imagen más natural, atributos que cada vez más toman en cuenta los consumidores. Este estudio tuvo como objetivo caracterizar nutricionalmente la carne de cordero, proveniente de cinco tipos genéticos, criados en un sistema pastoril, a través del contenido de minerales esenciales; macroelementos, Ca, Mg, Na y K, minerales traza como Se, Co, Zn, Cu, Mn, hierro total (TFe), hierro heme (HFe) y hierro no heme (NHFe) y vitamina B12 en el músculo *longissimus dorsi*. Se estudiaron lgenotipos Corriedale, Merino Dohne, Highlander, Corriedale Pro y la crusa Merino Australiano x Corriedale; n=10. El biotipo Merino Dohne tuvo la mayor concentración de calcio ($66,6 \pm 6,3 \text{ mg} \cdot \text{kg}^{-1}$), Highlander® y Merino Dohne tienen una concentración de manganeso significativamente ($P < 0,05$) mayor ($304,1 \pm 26,0$ y $308,7 \pm 23,6 \text{ } \mu\text{g} \cdot \text{kg}^{-1}$, respectivamente) que los demás genotipos. No hubo diferencias significativas en las concentraciones de vitamina B₁₂ entre los biotipos de corderos. La relación HFe y HFe/TFe fue mayor ($P < 0,05$) en los biotipos Corriedale y Corriedale Pro ($15,7 \pm 0,6$ y $15,4 \pm 0,7 \text{ mg} \cdot \text{kg}^{-1}$ y $81,7 \pm 2,8\%$ y $76,0 \pm 2,2\%$, respectivamente) y, en consecuencia, menor NHFe, en relación con los otros grupos. También se obtuvo un mayor contenido de Zn en Corriedale ($32,6 \pm 1,3 \text{ mg} \cdot \text{kg}^{-1}$), pero los otros biotipos también son ricos en zinc. Estos resultados demuestran que la carne de cordero de estos biotipos constituye una buena fuente para personas con altos requerimientos como niños y ancianos.

Palabras clave: carne de cordero, calidad de carne; minerales; hierro hemínico y no hemínico

4. Macrominerals, trace elements and hem and non-hem iron status in muscle Longissimus dorsi, from five double purpose lambs breed reared on pasture system in Uruguay

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4.1. Abstract

Sheep meat production is facing new challenges, so a thorough knowledge of the attributes of lamb meat produced by different genotypes and under pasture conditions is necessary to characterize these systems, to valorize and differentiate the product from a quality approach and towards a more natural image, attributes that are increasingly taken into account by consumers. This study aimed to characterize the lamb meat nutritionally, coming from five genetic types, reared in a pastoral system, through the content of essential minerals, macro element, Ca, Mg, Na and K, trace elements as Se, Co, Zn, Cu, Mn, total iron (TFe), hem iron (HFe) and non-hem iron (NHFe) and B12 vitamin in the Longissimus dorsi muscle. The breeds, Corriedale, Merino Dohne, Highlander®, Corriedale Pro, and Australian Merino x Corriedale crossbreed; n=10, were studied. Merino Dohne breed has the highest calcium concentration (66.6 ± 6.3 mg.kg⁻¹), Highlander® and Merino Dohne have a significantly ($P < 0.05$) higher manganese concentration (304.1 ± 26.0 and 308.7 ± 23.6 µg.kg⁻¹ g, respectively) than the other breeds. There were no significant differences in vitamin B12 concentrations between lamb breeds. The HFe and HFe/TFe ratio was higher ($P < 0.05$) in the Corriedale and Corriedale Pro breeds (15.7 ± 0.6 and 15.4 ± 0.7 mg.kg⁻¹ and $81.7 \pm 2.8\%$ and $76.0 \pm 2.2\%$, respectively) and consequently, less NHFe, related to others groups. Also, increased Zn content was obtained in Corriedale (32.6 ± 1.3 mg.kg⁻¹), but other breeds are also rich in zinc. These results show that meat from these biotypes qualifies as a good source claim for people with high requirements, such as children and elders.

Key words: Lamb meat; meat quality; minerals; haem and no haem iron

4.2. Introduction

Animal protein demand globally, driven by increasing population and discretionary income, is associated with better life quality in great Cities, fast information exchanges, marketing, and cultural evolution [1]. Throughout the world, different animal production systems coexist, with lamb production is the most extended and adapted to different regions, with many other breeds, and also with a predominance of the farm family system [2]. Lamb meat production in Uruguay has undergone various stages in the last five years, with a stable sheep stock of 6.2 million heads [3]. The primary breed reared in Uruguay was Corriedale (42%), a dual-purpose breed, and Australian Merino (26%) for wool production [3].

Due to the variability of external market demand and competitive prices, the Country has recently emphasized increasing the added value of products to be more competitive in international markets. Traditionally, sheep production in the Country is carried out in pastoral systems, associated with an image of animal welfare and natural products, whose exploitation is essential for export promotion and to offer healthy food for consumers [5]. A Region in the coastal Northwest of the Country, with abundant high-quality grasses in natural conditions, is traditionally lamb producer, mainly small-scale and predominately Merino for the wool [6]. Red meat is highly nutritional and has a high biological value of protein, bioavailable iron, trace minerals, and vitamins, including a high content of B12 [7]. Considering the pastoral-based systems, it observed that the genetic type [8], as well as the type of muscle and the age of the animal [9], impact some nutritional components, which can vary, as well as their oxidative and antioxidant potential [7, 10]. These differences could associate some racial cross with a specific nutritional composition to a particular oxidation potential and antioxidant capacity of the meat. Knowledge of the attributes of lamb meat produced by different genotypes in grazing conditions is necessary to characterize this meat and to add value, take into account the pastoral system, to contribute to human nutrition and consumer demand, and with the objectives of developing sustainable (SDO) for this Region of South America. This study aimed to characterize the meat nutritionally, coming from five genetic types, Corriedale, Corriedale Pro, Merino Dohne, Highlander, and Australian Merino x Corriedale cross breed, reared on a

pastoral system, through the content of essential minerals, macro element, Ca, Mg, Na and K, trace elements as, Se, Co, Zn, Cu, Mn and total iron, hem iron and non-hem iron in the *Longissimus dorsi* muscle.

4.3. Material and Methods

4.3.1. Animals

The study was carried out with lambs from the Experimental Station Mario Alberto Cassinoni (EEMAC) of the Faculty of Agronomy (Udelar) in Paysandú, Uruguay. Five genetic biotypes were studied: Corriedale, Merino Dohne, Highlander, Corriedale Pro, and Australian Merino x Corriedale crossbreed, n=10 in each group (TABLE I). The lambs were maintained in a single flock for the experiment, grazing forage. A strategic health control of lamb plan followed, and no lamb presented health problems during the study period, and no lamb presented health problems during the study period.

TABLE I
Live weight and age at slaughter of lambs for Corriedale, Corriedale Pro, Highlander, Merino Dohne and a Crossbreed (MA x C; Australian Merino x Corriedale), raising on pastures

Genotypes	Age at slaughter(days)	Live weight (kg)
Corriedale	339.8±4.7	49.3±5.3
Corriedale Pro	343.7±7.3	45.7±2.7
Highlander	340.4±5.4	53.0±3.6
Merino Dohne	341.3±5.5	55.5±3.8
Crossbreed (MAxC)	334.1±11.2	46.6±8.0

Data represent mean ± SEM of n=10 for each one of genotypes

4.3.2. Ethics statement

Animals used in this study were maintained in the facilities and environment of the Experimental Station of the Faculty of Agronomy (Udelar) in Paysandú, Uruguay, following the regulations of the University's ethics committee, the Honorary Commission for Animal Experimentation (CHEA, Udelar). The protocol used (Nº

1401) in this investigation was approved by the Ethical Commission for the Use of Animals (CEUA, CENUR Litoral Norte) following the regulations of the CHEA.

4.3.3. Experimental diets

Lambs were grazing on mixed pasture, including cocksfoot (*Dactylis glomerata*) and white clover (*Trifolium repens*.*l*) (available forage 2756 kgDM.ha-1) and on a winter annual crops oats (*Avena sativa*) in a rotational grazing with the availability of forage of 2743 kgDM.ha-1, as shown in TABLE II. The sample sward-cutting techniques and Botanal [11] were used to determine the amount of forage available and the types of vegetation present in the grazing area.

4.3.4. Muscles samples

At 72 h post mortem, the Longissimus dorsi muscle was excised from the carcass. Subcutaneous fat and silver skin (epimysium), was removed and packed in a vacuum (Vacuum Sealer Machine: Lacor, Model: 69050, Spain) and immediately transported to the laboratory in a cooler with ice packs. The samples were lyophilized (LGJ-12 Freezer Dryer, China) for minerals and non-hem determination. The samples were immediately used for haem iron analysis. Preparation of solutions and standards. Sub-boiling distilled HNO₃ 1 M, prepared with HNO₃ 65%, puriss. p.a. (84378, Merck, Germany); HCL 6 M, prepared with HCl 37%, EMSURE, puriss. p.a. (30721, Merck, Germany); Mg (NO₃)₂, in 17%HNO₃, magnesium matrix modifier 1% (63043, puriss. p.a. for graphite furnace-AAS, Fluka, Chemika, Switzerland); and Pd (NO₃)₂, in 15% HNO₃, palladium nitrate modifier 1% (B0190635, puriss. p.a. for graphite furnace-AAS, Perkin Elmer, Germany) was used for sample preparation and analysis. Millipore-Milli Q distilled deionized water (Merck KGaA, Darmstadt, Germany), with a resistivity of 18 MΩ cm⁻¹, was used throughout. Glassware was soaked for 24 h in dilute (50 mL.L⁻¹) distilled nitric acid and then rinsed thoroughly in distilled deionized water.

Sample Preparation. Samples of pasture (1 g, from a larger sample previously dried to 50°C for 48 h and grounded) and a sample of Longissimus dorsi muscle (10 g

previously freeze-dried) were dried in a forced-air oven at $105\pm2^{\circ}\text{C}$ (Labotecgroup, BJPX-Juneau, Uruguay) until the weight was constant. Subsequently, the samples were ashed in a covered crucible at 550°C in a muffle furnace (Thermolyne, Cimarec 3, USA) with a temperature ramp for 16 h to obtain white residual ash. The ashes were subjected to an acid digestion process in an Erlenmeyer flask, covered with a micro glass ball, with 1 M HNO₃ and 6 M HCl on a hot plate ($<80^{\circ}\text{C}$, Thermolyne, 48000 Furnace, USA), then filtered with ashless filter paper (Macherey-Nagel MN 640 d, Germany) and diluted to 10- or 25-mL final volume with distilled deionized water [12]. Blank was also prepared in the same procedure without a sample.

For Se and Co determination in pasture and meat samples, calibration solutions of Se (0, 35, 70, 140, and 280 $\mu\text{gSe.L}^{-1}$) were prepared immediately before use by dilution (with 0.2% distilled HNO₃ 65% in distilled and deionized water) of a 1000 $\mu\text{gSe.L}^{-1}$, HNO₃ 2% standard solution for AA (certified, N93000149, Perkin Elmer, USA). For Co measurements, calibration solutions (0, 5, 10, 20 $\mu\text{gCo.L}^{-1}$) were prepared immediately before use by dilution (with 0.2% distilled HNO₃ 65% in distilled and deionized water) of a 1000 $\mu\text{gCo.L}^{-1}$, HNO₃ 2% standard solution for AA (certified, N93000149, Perkin Elmer, USA).

Se and Co measurements in acidic aqueous dilution (blanks, samples, and calibration curve) were performed with an atomic absorption spectrophotometer (AAS Perkin Elmer, Analyst 300, USA) equipped with a deuterium lamp background corrector, a Perkin Elmer HGA 800, USA, graphite furnace, and a Perkin Elmer AS-800 autosampler, USA [13,14]. The determinations were conducted using matrix modifiers based on magnesium nitrate and palladium nitrate [15]. Argon (99% purity) was used as a carrier gas, and a selenium or cobalt HCL lamp was used as a light source. All determinations were performed in triplicate.

TABLE II

Macro and trace mineral content (mean ± SEM) in the mixed pasture (1) and oat, a winter annual grass (2) offered to five genotypes of lambs for the finishing period

Pastures	Available forage kg.ha ⁻¹	Minerals									
		Fe	Zn	Cu	Mn	Se	Co	Ca	Mg	K	
		mg.kg ⁻¹ DM ^b				µg.kg ⁻¹ DM		g.100 g ⁻¹ DM			
<i>Pasture 1</i>											
<i>Dactylis glomerata^a</i>	225.01± 50.72	10.45± 0.36	14.41± 0.76	114.11± 21.71	96.15 ± 13.37	49.40 ± 4.12	0.41± 0.03	0.15± 0.01	0.06± 0.00	1.22± 0.01	
<i>Trifolium repens^a</i>	2756	173.30± 25.77	12.78± 0.47	12.47± 2.10	78.86± 6.88	46.56± 5.38	122.46± 14.79	1.34± 0.12	0.37± 0.01	0.22± 0.04	1.32± 0.02
Weeds ^a		384.55	18.38	19.30	126.74	39.31	58.97	1.48	0.18	0.10	0.93
<i>Pasture 2</i>											
<i>Avena sativa</i>	2743	116.02± 17.29	6.2± 0.58	4.26± 0.52	71.70± 15.84	74.31± 16.66	90.05± 9.99	0.35± 0.03	0.14± 0.01	0.17± 0.02	1.23± 0.02

^a Botanical species mainly represented in mixed pasture 1 ^b DM= dry matter

The detection limit was calculated as three standard deviations of blanks/average of 10 blanks [15], and precision was calculated as RSD, %, of 10 measures.

Ca, Mg, K, Na, Fe, Zn, Cu, and Mn measurements in acidic aqueous dilution (blanks, samples, and calibration curve) were performed with an atomic absorption spectrophotometer (AAS Perkin Elmer, Analyst 300, USA) with flame, as described Jorhem et al. [16]. All samples were analyzed in triplicate.

After extraction with acidified acetone solution, total haem pigments in meat samples were determined as hemin [17]. Hemin was quantified by its absorption peak at 640 nm. Briefly, fresh meat samples (1 g) were finely chopped and macerated in 4.5 mL of 90% acidified acetone in 15x90 mm glass test tubes for 1 min on reduced light to minimize pigment fading during the extraction. The tubes were vortexed (ST-100, MRC, Israel), sealed to minimize evaporation, held at room temperature in the darkness for 1 hour. and then filtered with glass filter paper (Whatman® glass microfiber filters, Grade GF/A, Merck KGaA Germany). The haem iron content was calculated with the factor 0.0882 g iron/g hematin. All samples were assayed in duplicate.

The ferrozine method was the key in determining the non-haem iron [18]. Briefly, freeze-dried samples of meat (500 mg) were grounded using a mortar and pestle, dissolved in a mixture of 3 mL of 0.1 M citrate phosphate buffer (pH 5.5) and 1 mL of 2% ascorbic acid (as reducing agent) in 0.2 M HCl and left to stand at room temperature for 15 min before adding 2 mL of 11.3% trichloroacetic acid. After centrifugation at 3000 G for 10 min, the supernatant was recuperated. Reagents were added to 2 mL of the supernatant plus 0.8 mL of 10% ammonium acetate and 0.2 mL of ferrozine, and the absorbance was measured at 560 nm against a standard curve. All samples were assayed in duplicate.

From values obtained for cobalt (Co) content in muscle, vitamin B12 was calculated [19].

4.3.5. Statistical analysis

Data of all variables measured were presented as mean \pm standard error of the mean (SEM). The normality study of the variables was performed using the Shapiro-

Wills test. To determine the effect of breeds on the variables studied, a one-way ANOVA analysis was used for normally distributed variables and Kruskal Wallis test for non-normally distributed variables, following the mathematical model:

$$Y = \mu + T_i + e_j$$

where μ : mathematical average; T : treatment relative effect; e : experimental error

Y = response variable (macro and trace minerals, vitamin B12, hem iron and non-hem iron); i = lamb bread; j : random variable

When the lamb breed effect detected a significant difference, post hoc means multiple comparisons were realized by the Tukey and Kramer test at $P<0.05$ (normal distributed) and Wilcoxon test with (non-normally distributed).

To determine if it is possible to differentiate meat samples of breed based on their mineral content and gain further insight into the variables that have the most impact on lamb meat, the trace mineral composition dataset and iron forms, such as iron hem, iron non-hem and the ratio iron hem/total iron was included in the principal component analysis (PCA). Statistical analysis was carried out using the R software [20].

4.4. Results and Discussion

Our study on the mineral composition of mixed pasture and oats (*Avena sativa*) (TABLE II), reveals that the levels of Ca, Mg, Na, and K could potentially be inadequate for the requirements of different breeds of lambs Masters et al. [21]. This is a significant finding, considering that levels and bioavailability change with the season in the Southern Hemisphere Pittaluga [22] and that these requirements are not accurately determined in all breeds [21].

Following the requirements of the different genotypes studied here, the pasture's copper content is under the recommended levels according to the lambs under study [23]. Soils in Uruguay are widely deficient in copper, impacting hypocupremia in cattle (*Bos taurus*) in the coastal west region [24]. However, copper content in plants depends on botanical species, as shown in TABLE II, where cultivated oats are deficient in copper (< 10 mg.kg⁻¹ McDowell et al. [25]) and native pastures, legumes, and grasses are adequate (> 10 mg.kg⁻¹). Also, season and fertilization can affect the level of copper in native grass and cultivated pastures [22,26]. Suboptimal content in selenium in pasture mixed received in this study was observed previously by Guerra et al. [6] related to season, particularly in the coastal Region where this study was carried out. Still, it could change with the season, as reported before [6]. Cobalt in pastures fed by lambs it seems adequate to the requirements of lambs (> 0.1 mg.kg⁻¹ Masters et al. [21]). Concerning Fe and Mn in pastures, levels were higher than critical levels (>50 and >40 mg.kg⁻¹, respectively, McDowell et al. [25]), and it is not a problem for lambs. However, for zinc, pasture content is under the critical level of 30 mg.kg⁻¹ DM and thus does not fill the requirements of this mineral for growing lamb following the National Research Council recommendation (NRC) [23], but taking into account that relevant differences have been reported for different breeds [27,28], caution is due to conclude.

Macrominerals and trace minerals content in *Longissimus dorsi* meat from five genotypes (breed) were studied and raised on the pasture, as analyzed in TABLES III and IV.

Only Ca showed significant differences ($P<0.05$) among the macro minerals between breeds. Merino Dohne had the highest value (66.6 ± 6.3 mg.kg⁻¹), and the

lowest value was from Corriedale Pro (37.9 ± 5.4 mg.kg⁻¹). Williams [29], Purchas et al. [30], and Kasap et al. [31] reported Ca values between 4-11 mg.100 g⁻¹ of fresh meat. In contrast, Balhaj et al. [32] reported much higher values (between 41.96 - 59 mg.100 g⁻¹ fresh meat) than those found in this and other studies reported in the literature. This difference may be due to breed studied, muscle type, genre, body weight or feeding intensity Bellof et al. [33] since in that work, the lambs, German Merino received barley and mineral salt supplementation from weaning onwards, contrary to our work that lambs were only on pasture.

Animal muscle content calcium is not claimed to be a good source for humans but is essential to biochemical function, particularly for muscular fiber contraction [34]. However, the minimum content of calcium that muscles need to work and the meat quality in each lamb breed, need to be clarified, and the literature on this subject is scarce.

There were no significant differences in magnesium (Mg) ($P > 0.05$) between the different sheep breeds studied here being similar to values reported by Kasap et al. [31] and Hoke et al. [35]. There was no significant difference in sodium (Na) and potassium (K), being the values obtained were the same as those reported by Hoke et al. [35], Williams et al. [29], while Kasap et al. [31] said lower values than in this work.

Meat is a good source of vitamin B12 for humans [34]. The synthesis depends on cobalt as the primary component of vitamin B12, either as a cofactor for enzymes that require the vitamin or for microorganisms that synthesize the vitamin as a secondary metabolite. Ruminants need cobalt to provide to the rumen population of methanogenic bacteria synthesizing vitamin B12 [36,37]. Vitamin B12 has a molar mass of 1355 g.mol⁻¹ Suttle [38] and contains 4.4% cobalt [19,38]. Based on this ratio, the value of vitamin B12 was estimated in this study (TABLE III). There was no significant difference ($P > 0.05$) between sheep breeds in cobalt and vitamin B12 concentration. The concentrations obtained in this study are slightly lower than those reported by Juárez et al. [39] (0.6-2.5 µg.100g⁻¹). In sheep the efficiency of incorporation of cobalt in the molecule of vitamin B12 is lower than in bovine animals [37].

Copper, zinc, iron, and manganese are cofactors in several enzymes and contribute to the functioning of the immune system [40]. Iron is present in myoglobin and haemoglobin, proteins responsible for oxygen transport in the blood [41].

TABLE III

Macro, trace mineral, and calculated vitamin B₁₂^(a), in raw *Longissimus dorsi* from lamb's breeds (Corriedale, Corriedale Pro, Highlander, Merino Dohne) and one crossbreed (Australian Merino x Corriedale, MAxC) double purposes reared on pasture

Genotypes	Minerals						Vitamin			
	Ca	Mg	Na	K	Zn	Cu	Mn	Se	Co	
	mg.kg ⁻¹						μg.kg ⁻¹			ng.g ⁻¹
Corriedale	50.5± 7.5 ^{ab}	244.7± 12.4	719.9± 52.7	3572.1± 205.3	32.6± 1.3 ^a	1.40± 0.13	251.9± 34.4 ^{ab}	91.1± 7.3	28.6± 3.7	1.26± 0.17
Corriedale Pro	37.9± 5.4 ^b	269.0± 3.1	792.3± 29.9	3846.4± 51.2	29.6± 0.7 ^{ab}	1.57± 0.10	281.9± 33.8 ^{ab}	81.7± 3.8	37.5± 5.3	1.65± 0.23
Highlander®	64.1± 7.0 ^{ab}	262.9± 2.7	694.7± 29.9	3826.5± 54.6	30.9± 0.9 ^{ab}	1.50± 0.09	304.1± 26.0 ^a	86.2± 6.7	26.8± 3.7	1.18± 0.17
Merino Dohne	66.6± 6.3 ^a	258.1± 6.8	708.6± 31.1	3689.3± 86.5	31.4± 0.9 ^{ab}	1.46± 0.14	308.7± 23.6 ^a	92.2± 2.8	29.1± 3.5	1.28± 0.15
Crossbreed (MA x C)	44.2± 6.7 ^{ab}	258.4± 4.1	669.3± 32.1	3569.4± 60.4	27.9± 0.7 ^b	1.46± 0.09	191.5± 14.7 ^b	76.7± 7.9	34.5± 3.8	1.52± 0.17
P value	0.01	n.s.	n.s.	n.s.	0.03	n.s.	0.027	n.s.	n.s.	n.s.

^(a) Calculated in base to Girard *et al.* (2009). Data are presented as mean ± SEM of n=10. ^{a,b} Data in each column with different lowercase letters show a significant difference between breeds or crossbreed, P<0.05.

Highlander® and Dohne Merino sheep breeds have significantly higher manganese concentrations (304.1 ± 26.0 and $308.7 \pm 23.6 \text{ } \mu\text{g}.\text{kg}^{-1}$, respectively) ($P < 0.05$). These breeds show high values (60%) concerning those presented by the crossbreed (MA x C), although they were not statistically significantly different from Corriedale. Only some studies have reported Mn values in lambs. here are only a few studies that have reported Mn levels in lambs. In studies by Hoke et al. [35], values of $14 \text{ } \mu\text{g}.100 \text{ g}^{-1}$ of lean meat were reported for the Loin Chop Cut.

There was no difference in copper levels between different sheep breeds of lambs, even though copper was at a critical level in the pasture. Copper concentrations were similar to those of Lombardi-Boccia et al. [42] and Juárez et al. [39]. Selenium in the meat of five breeds is adequate for nutrition children, ranging from $76.7 \text{ } \mu\text{g}.\text{kg}^{-1}$ to $91.1 \text{ } \mu\text{g}.\text{kg}^{-1}$. Indeed, 100 grams of this raw lamb meat contributes of 20-25 % of RDA [14].

The Longissimus dorsi muscle of the Corriedale sheep breed presented a higher concentration of zinc ($32.6 \pm 1.3 \text{ mg}.\text{kg}^{-1}$) than the other sheep breeds and crossbreeds ($P < 0.05$). Australian studies across different lamb production systems reported zinc levels on average of $2.43 \text{ mg}.100 \text{ g}^{-1}$ of muscle Mortimer et al. [43], Pannier et al. [44], 34% lower than in the present investigation. Zinc is the second most deficient mineral in many countries [45]. Its role has been actualized related to antiviral immunity Read et al. [46], and this meat contributes mainly to the recommendations to protect children and older people Saadoun et al. [47] by increasing immune defenses, among other beneficial effects.

Corriedale and Corriedale Pro sheep breeds show significantly more Hem Fe and a higher ratio of HFe/TFe, but no difference for Total Fe was obtained (TABLE IV). These results are interesting because iron is a relevant mineral in human nutrition, particularly in children, pregnant women, and adolescents [48]. Iron dietary deficiency is a severe health problem affecting 20- 39 % of children worldwide (data from the World Health Organization (WHO) [49]). The diet has two types of iron: hem iron, derived from haemoglobin, and myoglobin, which represents a small fraction of total iron and is well absorbed, and non-hem iron, inorganic iron, with low availability. Hem iron is provided by animal proteins such as meat [41].

TABLE IV

Moisture, ashes content, total iron (TFe), hem (HFe) and non-hem iron (NHFe) and the ratio HFe/TFe (%), in raw *Longissimus dorsi* from lamb's breeds (Corriedale, Corriedale Pro, Highlander, Merino Dohne) and one crossbreed (Merino Australian x Corriedale, MAxC) double purposes reared on pasture

Genotypes	Moisture	Ashes	TFe	HFe	NH Fe	HFe/TFe
	g.100 g ⁻¹			mg.kg-1		
Corriedale	75.6±2.7	0.90±0.01	19.5±0.9	15.7±0.6 ^a	3.6±0.6 ^b	81.7±2.8 ^a
Corriedale Pro	75.3±2.9	0.85±0.02	20.2±0.6	15.4±0.7 ^{ab}	4.6±0.5 ^b	76.0±2.2 ^{ab}
Highlander	74.9±2.6	0.85±0.02	20.2±1.9	13.7±0.4 ^{ab}	6.4±0.5 ^a	68.2±2.1 ^b
Merino Dohne	75.1±2.8	0.87±0.02	20.5±0.6	13.3±0.6 ^b	7.1±0.9 ^a	65.7±3.7 ^b
Crossbreed (MAxC)	75.2±2.8	0.87±0.02	20.7±0.8	14.5±0.7 ^{ab}	6.3±0.3 ^a	70.2±1.2 ^b
P value	n.s	n.s.	n.s	0.027	0.001	0.001

Data are presented as mean SEM ± of n=10. ^{a, b}. Data in each column with different lowercase letter show significant difference between breeds or crossbreed, P<0.05.

There was no statistical difference in total iron (TFe) concentration between the five breeds (TABLE IV). Pannier et al. [50], in a study based on 2,000 lambs and three main genotypes, did not observe differences in TFe, but the difference between Zn levels was observed in different sheep breeds. This response coincides with what was obtained in our work. On the other hand, there was a significant difference ($P<0.05$) in the content of hem iron (HFe) and the ratio HFe/TFe between sheep breeds. Corriedale was the sheep breed with the highest hem iron (HFe) 15.7 ± 0.6 mg.kg⁻¹ and HFe/TFe (81.7 %) than Merino Dohne sheep breed (13.3 ± 0.6 mg.kg⁻¹ and 65.7 %). Although the difference in absolute value is 2 mg between the two breeds, this represents a difference of 15%, so it can be considered a significant difference. As Hem iron is part of myoglobin, and the concentration of myoglobin depends on the type of muscle fiber, being higher in oxidative type fiber (Type I and IIa), red fibers, and lower in glycolytic type fiber (Type IIx, IIb) in white muscle fiber [51, 52, 53]. A possible explanation is that sheep breeds studied here present differences in fiber type at the same slaughter age [53]. Cottle [54] reported that the Merino Dohne sheep breed could have fibers type IIb (IIx) related to a leaner carcass; consequently, a lower myoglobin content is possible and, therefore, less hem iron. Previous studies by Pannier et al. [44] reported a positive association between aerobic markers and mineral content as iron but in lesser magnitude with zinc.

4.4.1. Principal component analysis of meat quality

Principal component analysis (PCA) does carry out to show the relationships among meat trace minerals and iron forms of five breeds produced in Uruguay (FIG. 1). The result of this analysis indicates the genotype effect influenced the majority of the parameters. The PCA makes it possible to visualize a large number on a single graph and estimate the statistical links between the studied individuals.

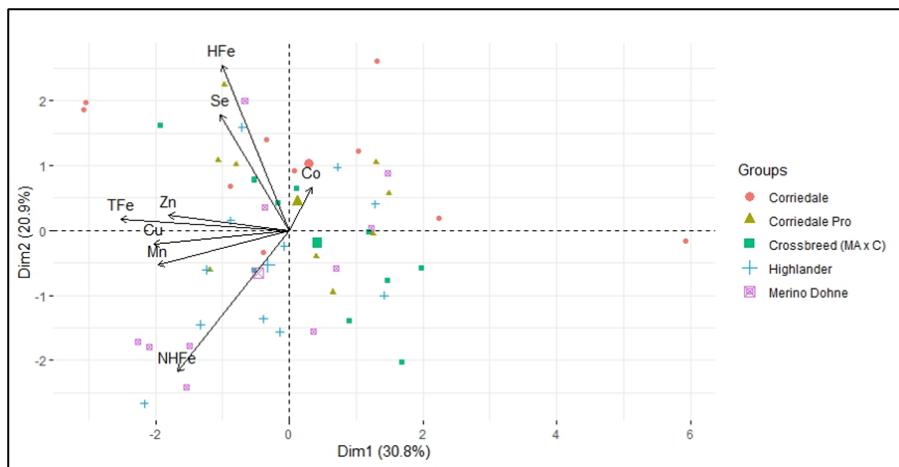


FIGURE 1. Map of selected minerals trace and iron forms of lamb meat from five lamb breeds and cross-breed groups reared on pasture

The loading values for principal component one (PC1) show a strong and positive association with TFe (0.833), Cu (0.670), Mn (0.649) and Zn (0.598). Loading values in PC2 are high and positive for HFe (0.844) and Se (0.591) and negative for NHFe (-0.718). In summary, the variables that best represent PC1 are TFe (28.14%) and Cu (18.23%), while HFe (42.58%) and NHFe (30.86%) are the variables that contribute most to PC2. Co is the mineral trace that is poorly represented for both principal components.

The individual projection shows no clear discrimination between the breeds studied. However, an association can be observed between the Merino Donhe and Highlander® breeds, which in turn, are highly associated with the mineral HNFe.

Based on PCA statistical analysis, we identified variations in the trace mineral content of the meat among the different animals studied. If the principal components PC1 and PC2 are analysed together, TFe and its components (HFe and NHFe) are the variables that most account for the original variability. All in all, however, there is considerable heterogeneity in the distribution or variability of dispersion of the genotypes studied (which somewhat lowers the level of precision, reliability, or accuracy of the groups formed).

Meat mineral trace content, principally iron and iron components raised on pastures, is an excellent way to satisfy the requirements of human people. If one wants

to increase the value of the meat market, it is crucial to consider the differences between sheep breeds, especially those bred for meat and wool production.

4.5. Conclusion

In the study context, the genetic component would not be a determining differential for the content of macrominerals and trace elements.

The sheep genotype studied showed a significant association with the minerals Mn, Ca, and Zn, as well as the quantities of HFe and NHFe.

. This study justifies and gives excellent scope for further research on sheep breeds and possible interactions between factors such as age, slaughter weight, different diets, and their effect on meat and wool quality.

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Disclosure statement

The authors declare no conflict of interest.

Data availability statement

Data is available on request from the authors. The data that support the findings of this study are available from the corresponding author, Guerra MH, upon reasonable request

Conflict of interest

The authors declare that they have no conflict of interest.

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Ácidos grasos, índices lipídicos de salud y actividades enzimáticas en el músculo *longissimus thoracis*, de seis razas de corderos producidos sobre pasturas en el norte de Uruguay

Resumen

Se determinó la composición en ácidos grasos de los glicerolípidos y glicerofosfolípidos del músculo *longissimus thoracis* de seis biotipos de corderos producidos con pasturas en Uruguay. También se cuantificaron los ácidos grasos de cadena ramificada monometiles iso y anteiso, y el contenido de ácidos grasos impares de la carne. Se determinaron índices lipídicos de salud y actividades de las enzimas del metabolismo de los ácidos grasos. Los corderos estudiados fueron machos de 11-12 meses de biotipos Highlander®, (H), Merino Dohne (MD), Corriedale (C), Corriedale Pro, (CPRO), un cruce entre Corriedale X Australian Merino (CxAM) y Romney Marsh (RM). Los animales fueron criados sobre pasturas en condiciones idénticas sin suplementos. El pastoreo fue rotativo basado en una avena de cultivos anuales de invierno (*Avena sativa* spp.), cocksfoot, (*Dactylis glomerata* spp.) y trébol blanco (*Trifolium repens* spp.). Los resultados no mostraron diferencias sustanciales entre biotipos en la composición en ácidos grasos de la carne, excepto por ácidos grasos relevantes como C16:0 (MD, C), C18:3n3 (H, C) y CLA (H, CPRO, CxAM) para glicerolípidos. También C18:1 (H, CPRO y CxAM), C18:2n6 (H, CxAM) y C18:3n3 (H, C) para glicerofosfolípidos. Asimismo, hay otras diferencias como el contenido de ácidos grasos anteiso (RM, MD) y la relación del índice hipocolesterolémico/hipercolesterolémico (MD, C). Para las actividades enzimáticas del metabolismo de los ácidos grasos, el MD mostró una menor enzima desaturasa Δ-9 para C16:0 que C, CPRO y CxAM. Además, H mostró una menor actividad de la enzima Δ-6 desaturasa que C, y tanto MD como CxAM mostraron una menor actividad de la enzima elongasa que C. Los resultados mostraron que la carne de cordero de los diferentes biotipos presenta en general buenos indicadores nutricionales de lípidos, en comparación con los resultados de otras investigaciones en corderos. Esta información podría ayudar a los productores de corderos del Uruguay a promover sus productos sobre la base de datos científicos.

Palabras clave: carne de cordero; ácidos grasos; sistema extensivo; pastura

5. Fatty acids composition, lipids health indices and enzyme activities of longissimus thoracis muscle of six breeds of sheep produced on pasture in Northern region of Uruguay

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5.1. Abstract

The determination of the composition of fatty acids of glycerolipids and glycerophospholipids of meat from *longissimus thoracis* of six biotypes of lamb produced on pasture in Uruguay was undertaken of meat were quantified. Also, some lipid health indices and lipid metabolism enzymes were determined. The studied lambs were males aged 11-12 months of biotypes Highlander® (H), Merino Dohne (MD), Corriedale (C), Corriedale Pro (CPRO), a crossing between Corriedale X Australian Merino (CxAM) and Romney Marsh (RM). The animals were reared on pasture in identical conditions without supplementation. The grazing was rotational based on winter annual crops oats (*Avena sativa* spp.), cocksfoot (*Dactylis glomerata* spp.) and white clover (*Trifolium repens* spp.). The results of the study did not show substantial differences between breeds regarding the fatty acids composition of meat, except a few relevant fatty acids such as C16:0 (MD > C), C18:3n3 (H < C) and CLA (H < CPRO, CxAM) for glycerolipids. Also, C18:1 (H > CPRO, CxAM), C18:2n6 (H < CxAM) and C18:3n3 (H < C) for glycerophospholipids. Likewise, other differences were outlined such as the anteiso monomethyl fatty acid content (MD < RM), and the hypocholesterolemic/hypercholesterolemic ratio (MD < C). MD showed a lower Δ-9 desaturase enzyme for lipids metabolism enzymes indices C16:0 than C, CPRO and CxAM. Also, H showed a lower Δ-6 desaturase enzyme activity than C, and both MD and CxAM showed a lower elongase enzyme activity than C. The results of the presents investigation showed that the meat of the lamb of the different breeds overall present suitable lipids nutritional indicators, in comparison with the results of other research in lambs. That information could help lamb producers in Uruguay to promote their products based on scientific data.

Key words: lamb meat; fatty acids; extensive system; pasture.

5.2. Introduction

Sheep (*Ovis aries*) meat has been a valuable food for human nutrition for thousands of years [1]. Sheep meat is available in many countries, often produced and consumed locally. Approximately 82 % of sheep breeds in the world are local breeds well adapted to their particular biotope, most of them fed local pasture [2]. However, commercial breeds are also the basis of the international sheep meat trade, often associated with the wool trade. Sheep meat provides consumers with protein, lipids, minerals (particularly iron and zinc), and vitamins, all of them necessary for adequate growth and metabolism function at all ages. Lipids are particularly relevant through their fatty acid composition since they are associated with some chronic diseases. Indeed, ruminant meat from sheep or lamb contains glycerolipids and glycerophospholipids composed of saturated fatty acids (SAT), monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA). Some SATs are associated with the occurrence of cardiovascular pathologies and cancer in human. Meanwhile MUFA seems to have beneficial effects on health [3]. For another part, PUFA, such as linoleic acid and α -linolenic acid, are essential for human nutrition and metabolism, meaning they must be present in the diet. The latter is a precursor for the biosynthesis of EPA (C20:5n3) and DHA (C22:6n3), two n-3 fatty acids involved in the protection against cardiovascular diseases and cancer in humans.

In Uruguay, sheep production is based on pasture and constitutes a relevant part of the meat market and the country's economic scheme. Various sheep breeds and crossings are present, and producers always been interested in improving their knowledge about the breeds they produce, mainly in genetic aspects linked to the wool quality. The primary breed present in Uruguay is the Corriedale (42 % of total sheep breeds) because of its dual-purpose characteristics in producing wool and meat. However, in the last few years, sheep producers' incomes in Uruguay have depended much more on meat, for both domestic and international trade, than wool. This is probably due to the relatively stable world market for sheep meat in comparison to the wool market and the positive future prospects for sheep meat trade in the region [4]. Consequently, producers are now interested in the nutritional quality of their sheep meat to help promote their products, mainly in the international sheep meat market.

Therefore, the present study has been undertaken to determine the composition of fatty acids in meat obtained from six breeds and crossings produced in Uruguay, including Corriedale, and fed exclusively with pasture. Some of those breeds have been recently introduced in the country, and scarce, or no information, to our knowledge, about the nutritional values of their meat could be sourced in the scientific literature. Furthermore, the study will generate information about some lipid health indices for consumers and fatty acid metabolism indices related to the enzyme desaturases, elongases and thioesterases.

5.3. Material and Methods

5.3.1. Animals and feeding

The meat studied in the work come from males of six breed produced in Uruguay on extensive system condition based on pasture. 1) Highlander® (H, n= 15; slaughtered at 54.38 ± 4.45 kg of body weight, 339 ± 6.7 days of age). H is a composite breed ($\frac{1}{2}$ Romney, $\frac{1}{4}$ Texeland $\frac{1}{4}$ Finnish Landrace) introduced in Uruguay on year 2005. 2) Merino Dohne (MD, n=11; slaughtered at 55.05 ± 3.72 kg of body weight, 341 ± 5.8 days of age), a dual-purpose breed originated in South Africa and introduced in Uruguay from Australia on year 2002. 3) Corriedale, (C, n=11; slaughtered at 50.3 ± 5.35 kg of body weight, 339 ± 4.83 days of age), a dual-purpose breed obtained by crossing Merino and Lincoln breeds in Australia and New Zealand around the years 1874-1880. C was introduced in Uruguay on 1916. 4) Corriedale Pro (CPRO, n= 15; slaughtered at 46.54 ± 5.53 kg of body weight, 341 ± 7.57 days of age), CPRO is a composite crossbreed developed in Uruguay, based on a crossing of Freisian Milchschaaf (25 %) with Finnish Landrace (25 %) and C (50 %). CPRO has been developed principally to improve the prolificacy without the loss of double purpose attribute of C. 5) A crossbreed between Corriedale and Australian Merino breed (CxAM, n= 15; slaughtered at 48.18 ± 7.04 kg of body weight, 334 ± 10.1 days of age). CxAM has been developed in Uruguay to improve the resistance to the gastrointestinal parasitism. 6) The last breed used in the study was Romney Marsh (RM, n=4; slaughtered at 48.92 ± 6.82 kg of body weight, 335 ± 2.5 days of age). RM is a dual-purpose breed, developed in England, and introduced in Uruguay on year 1896. Although only 4 animals RM were obtained from producers, the results of the experiment with those animals have been anyway included in the study, taking into account the long presence of that breed in the productive scheme of the country and the lack of nutritional information of RM meat produced in Uruguay.

Animals were maintained in the facilities of the Experimental Station of the Faculty of Agronomy (Udelar) in Paysandú - Uruguay, following the regulations of the University's ethics committee. The investigation has been approved by the Honorary Commission for Animal Experimentation (CHEA, Universidad de la

República, Udelar, Uruguay), recorded as protocol number 1401. Furthermore, the investigation has been also approved by the Ethical Commission for the Use of Animals (CEUA, CENUR, Udelar).

Animals have grazed pasture, without any supplementation, with a maximum animal density of 6 sheep by hectare, and rotated in paddocks of 15 hectares. The animals were reared on pasture, in groups separated by breed. Pasture (P1) consisted by a winter annual crops oats (*Avena sativa* spp.) with the availability of forage of 2743 kg DM/ha, that pasture has been used in a rotational grazing. Furthermore, the lamb grazed another pasture (P2) principally constituted by cocksfoot, (*Dactylis glomerata* spp.) and white clover (*Trifolium repens* spp.) with an availability of forage of 2756 kg DM/ha. All groups have been concomitantly transferred between P1 and P2 and inversely, depending of the availability of forage. For the sampling and the estimation of available forage and botanical composition in the grazing area, the cutting method "Sample Sward-cutting techniques" and Botanal was used [5]. The lipids and fatty acid composition of pasture was presented in TABLE I.

TABLE I

Lipid content (% of dry matter) and fatty acids composition of glycerolipids and glycerophospholipids (g/100 g fatty acids) of pastures grazed by lambs

	Pasture P1		Pasture P2	
	Oat (<i>Avena sativa</i>)	Legumes (<i>Trifolium repens</i>)	Gramineae (<i>Dactylis glomerata</i>)	Undefined Pasture
Lipids	3.38 ± 0.03	2.10 ± 0.02	3.65 ± 0.03	3.25 ± 0.03
C14:0	1.47 ± 0.71	1.26 ± 0.79	0.61 ± 0.06	0.65 ± 0.13
C16:0	16.9 ± 0.69	13.8 ± 0.57	15.0 ± 0.42	18.7 ± 0.63
C16:1	1.49 ± 0.06	1.51 ± 0.06	2.64 ± 0.24	2.49 ± 0.05
C18:0	1.92 ± 0.02	2.07 ± 0.08	1.18 ± 0.11	2.08 ± 0.03
C18:1	2.92 ± 0.10	3.35 ± 0.56	1.79 ± 0.04	2.83 ± 0.09
C18:2n6	8.88 ± 0.36	22.4 ± 0.68	10.3 ± 0.10	13.4 ± 0.16
C18:3n3	49.2 ± 1.41	45.6 ± 1.06	60.5 ± 0.65	50.0 ± 1.18
C20:0	0.58 ± 0.13	1.80 ± 0.10	0.30 ± 0.01	0.95 ± 0.37
C22:0	2.51 ± 0.19	2.47 ± 0.78	0.81 ± 0.03	1.17 ± 0.07
C24:0	3.74 ± 0.20	0.78 ± 0.12	1.29 ± 0.07	0.98 ± 0.07
Unidentified fatty acid	10.4 ± 1.38	4.91 ± 0.66	5.67 ± 1.22	6.68 ± 0.34
C14:0	0.12 ± 0.02	0.17 ± 0.03	0.08 ± 0.01	0.12 ± 0.02
C16:0	16.4 ± 0.48	12.4 ± 1.40	13.2 ± 0.54	17.2 ± 0.55
C16:1	1.97 ± 0.25	1.94 ± 0.14	1.72 ± 0.22	3.18 ± 0.19
C18:0	1.10 ± 0.07	1.48 ± 0.12	0.88 ± 0.05	1.19 ± 0.01
C18:1	2.11 ± 0.04	2.42 ± 0.46	1.33 ± 0.06	2.36 ± 0.09
C18:2n6	7.06 ± 0.34	13.4 ± 1.05	7.28 ± 0.09	12.0 ± 0.04
C18:3n3	51.0 ± 0.80	54.5 ± 4.31	66.7 ± 1.15	54.4 ± 2.18
C20:0	0.48 ± 0.05	1.67 ± 0.71	0.31 ± 0.08	1.06 ± 0.45
C22:0	3.52 ± 0.24	4.56 ± 0.76	1.13 ± 0.15	1.59 ± 0.12
C24:0	4.52 ± 0.56	1.52 ± 0.11	1.65 ± 0.32	1.27 ± 0.19
Unidentified fatty acid	11.6 ± 1.41	5.94 ± 0.89	5.61 ± 1.25	5.60 ± 1.9

Data are mean ± SEM of three samples of pasture. Animals have been concomitantly transferred between P1 and P2 and inversely, depending of the availability of forage.

The lambs were slaughtered in a commercial slaughterhouse (Certified Food Standard, Grade A, Certification Body LSQA S.A. for exportation by BRC Global Standard). At 72 hours *post mortem* the *longissimus thoracis* muscle (between 9th and 12th vertebrate) was withdraw, vacuum packaged and stored at -20 °C, until analysed.

5.3.2. Analytical determination

The plant lipids were determined on a dry ground sample, dried at 105 °C for 6 hours in a forced air dryer. Lipids of three replicates of 10 g were extracted by Soxhlet method (AOAC Method 945.16), using hexane (Carlo Erba, France, HPLC grade) as extraction solvent. The intramuscular lipids were extracted according to Folch *et al.* [6]. Briefly, a sample of 4 grams of meat of *longissimus thoracis* muscle (free of dissectible visible fat) was homogenized at 25000 rpm with an IKA T25 homogenizer (IKA Brandt, Sweden) during 1 min with 80 mL of chloroform: methanol 2:1, (Baker brand HPLC grade, USA). Afterward, the homogenate was filtered on fritted funnel (Fisher brand, graduation M, USA), transferred to a separating funnel, mixed by shaking and inverting for one minute and decanted overnight. The lower phase (chloroform containing lipids) was recuperated in a glass balloon (Fisher Brand, USA), evaporated at 45 °C with a light vacuum in a Rotavapor (IKA basic, Sweden). The balloon was dried in an oven at 35 °C for 60 min and cooled at ambient temperature overnight in a vacuum desiccator protected from light. The balloon was weighted at 0.0001 g. to determine the percentage of lipids of each sample. The methylation of fatty acids from glycerolipids fraction followed the procedure described by Ichihara *et al.* [7]. That procedure targets the fatty acids from triacylglycerols as well as those from phospholipids, both associated to a glycerol backbone. For the selective methylation of fatty acids from glycerophospholipids (polar glycerolipids), the procedure described by Ichihara *et al.* [8] has been used. The determination of fatty acids by gas chromatography followed a procedure using fused-silica capillary column CPSIL-88 of 100 m installed in a split/split less chromatograph Clarus 500 (Perkin Elmer Instruments, USA) with a FID detector. The samples injection was done using an autosampler from the same manufacturer. One μ l of each methylated sample was injected with a split ratio of 50 %. Hydrogen (Brand Linden, Uruguay, purity of

99.9995 %) was used as carrier gas having a ratio air/H₂ of 350 mL/35 mL. Filtered air was proven by compressor GAST model 3HBB-11T-M300AX (USA). The thermal conditions were: Injector/detector temperatures 250 °C/250°C, oven held at 90°C for one minute after the injection of the sample. The split valve was open 30 seconds after injection. Afterward the oven temperature was increased to 225 °C at 15°C/min. Fatty acids methylated esters (FAMEs) were determined comparing the retention time of authentic standards and the 37 component FAME standard mixture (Sigma-Aldrich, USA). Individuals FAME were quantified as a percentage of total detected FAMEs. The integration of signal has been conducted on Total Chrome software from Perkin Elmer (USA).

5.3.3. Calculus of health indices

The calculus of health indices was performed from the data of fatty acid composition of glycerolipids. The following indices were calculated:

-Indice of atherogenicity (AI): Compute the relationship between the sum of the main saturated fatty acids (pro-atherogenic) and the unsaturated (anti-atherogenic) fatty acids [9]. It was calculated as follows:

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

-Indices of thrombogenicity (TI): Estimate the potential to form clots in the blood vessels [9], determined by the relationship between the pro-thrombogenic and the anti-thrombogenic fatty acids (Sum of MUFA and PUFA). It was calculated as follows

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

-Hypocholesterolemic/Hypercholesterolemic ratio (h/H): Compute the relation between unsaturated fatty acids (MUFA and PUFA) and the saturated fatty acids 14:0 and 16:0. The h/H ratio was calculated according to Fernández *et al.* [10] as follows:

$$\mathbf{h/H} = (\text{C14:1} + \text{C16:1} + \text{C18:1} + \text{C20:1} + \text{C22:1} + \text{C18:2} + \text{C18:3} + \text{C20:3} + \text{C20:4} + \text{C20:5} + \text{C22:4} + \text{C22:5} + \text{C22:6}) / (\text{C14:0} + \text{C16:0})$$

5.3.4. Enzyme activity indices

The enzyme activity of desaturases, elongase and thioesterase was estimated by relating the amount of the specific substrate to the corresponding product of the respective enzyme. The calculus of those indices was performed from the data of fatty acid composition of glycerolipids. The calculated ratios were 16:1n-7 to 16:0 and 18:1n9 to 18:0, and their sum, for the activity of stearoyl-CoA desaturase (Δ -9-desaturase). The Δ -5 desaturase and Δ -6 desaturase were used as an index for the estimation of catalyzing the formation of long chain n-6 and n-3 starting from the corresponding precursor C18:2n6 and C18:3n3, respectively. Also, the ratio 18:0 to 16:0 was calculated to estimate the elongase activity. The thioesterase was estimated as the ratio of C16:0 to C14:0. These indices can be used as surrogates of the measure of the true enzyme activities [11].

5.3.5. Statistical analysis

Data are presented as mean \pm SEM. Results were analyzed by ANOVA one way to compare six genotypes, and *post hoc* Tukey-Kramer Test ($P < 0.05$), using the NCSS 12 Statistical Software (NCSS 2018, LLC. Kaysville, Utah, USA, <https://www.ncss.com>).

In addition, a principal component analysis (PCA) on the standardized variables at unit scale, associated to human health as intramuscular fat content (lipids), and C14:0, C16:0, C17:0ai, C18:3n3, CLA, BCFAai and BCFAi of the total fatty acids

were conducted to evaluate the relative differences of the meat samples in these lipid profile among breeds.

Another principal component analysis (PCA) on the variables associated to structure of membrane as C16:0, C16:1, C18:1, C18:2n6, C18:3n3 C20:4n6, EPA, DPA, DHA in glycerophospholipids fraction to evaluate if the breeds present differences. Variables were graphed in a biplot with different color related their contribution and the distribution of observations were graphed in a biplot using ellipses with 95% confidence interval. Statistical analysis was conducted using the PCA function of the package FactoMineR for the principal components analysis in the R software version 4.2.2 (R Core Team, 2022). To visualize de PCA results, thefactoextra package in the R software was used.

5.4. Results and Discussion

5.4.1. Lipids

The intramuscular fat content is one of the most relevant parameters, principally through the composition of fatty acids, when the nutritional quality has to be considered to characterize ruminant meat. Thus, depending on their specific fatty acid composition, lipids in meat could positively or negatively affect consumers' health. Indeed, they could respond positively to the nutritional requirement for growth and metabolism at all ages of consumers. Still, they could negatively influence human health through cardiovascular and cancer diseases [3,12]. However, for most consumers, the content of lipids expressed as g of total lipids by 100 g of meat is perceived as a key indicator to classify meat products, and foods regarding their incidence on health.

In the present work, the comparison between the different breeds showed lipids contents ranged between 2.39-4.49 g of lipids by 100 g of meat. Significant differences have been observed only between CXAM and H (TABLE II).

TABLE II

Lipids content (% of wet tissue) and fatty acids composition (g/100 g fatty acids) of glycerolipids present in longissimus thoracis muscle from lambs of different breeds produced on pasture

	Breeds						P≤
	H (n=15)	MD (n=11)	C (n=11)	C PRO (n=15)	CxAM (n=15)	RM (n=4)	
Lipids	2.39b ± 0.18	3.21ab ± 0.44	3.46ab ± 0.30	4.17a ± 0.40	4.25a ± 0.33	4.49ab ± 1.01	0.009
Saturated Fatty acids (SAT)							
C14:0	2.52 ± 0.23	3.46 ± 0.36	2.43 ± 0.20	3.04 ± 0.34	3.10 ± 0.28	2.45 ± 0.47	NS
C15:0i	0.12 ± 0.01	0.14 ± 0.01	0.12 ± 0.01	0.13 ± 0.01	0.12 ± 0.01	0.12 ± 0.02	NS
C15:0ai	0.16 ± 0.01	0.17 ± 0.02	0.14 ± 0.01	0.16 ± 0.01	0.15 ± 0.01	0.14 ± 0.03	NS
C15:0	0.51 ± 0.04	0.59 ± 0.04	0.50 ± 0.03	0.55 ± 0.03	0.51 ± 0.04	0.46 ± 0.08	NS
C16:0i	0.16 ± 0.01	0.17 ± 0.01	0.17 ± 0.01	0.16 ± 0.01	0.15 ± 0.01	0.18 ± 0.03	NS
C16:0	24.0ab ± 1.03	26.1a ± 1.43	21.2b ± 0.78	23.0ab ± 0.89	24.4ab ± 0.92	21.1ab ± 0.62	0.02
C17:0i	0.48 ± 0.04	0.39 ± 0.05	0.49 ± 0.04	0.52 ± 0.03	0.48 ± 0.03	0.55 ± 0.05	NS
C17:0ai	0.47a ± 0.03	0.35b ± 0.06	0.51a ± 0.04	0.50a ± 0.03	0.52a ± 0.02	0.58a ± 0.02	0.01
C17:0	1.50ab ± 0.06	1.35b ± 0.08	1.67a ± 0.06	1.59ab ± 0.04	1.47ab ± 0.05	1.57ab ± 0.12	0.03
C18:0	20.4ab ± 0.72	18.9ab ± 0.33	21.1a ± 0.50	18.9ab ± 0.62	18.3b ± 0.62	20.8ab ± 0.97	0.01
C20:0	0.10 ± 0.01	0.10 ± 0.01	0.13 ± 0.01	0.13 ± 0.01	0.11 ± 0.01	0.12 ± 0.02	NS
C22:0	0.02 ± 0.01	0.05 ± 0.01	0.05 ± 0.01	0.05 ± 0.01	0.04 ± 0.01	0.05 ± 0.01	NS
ΣSAT	49.3 ± 1.08	50.9 ± 1.58	47.3 ± 0.92	47.4 ± 0.77	48.1 ± 1.03	47.1 ± 0.87	NS

	Breeds							
	H (n=15)	MD (n=11)	C (n=11)	C PRO (n=15)	CxAM (n=15)	RM (n=4)	P≤	
C16:1	1.77ab ± 0.11	1.57b ± 0.10	1.80ab ± 0.09	2.04a ± 0.07	2.10a ± 0.11	1.87ab ± 0.11	0.004	
C17:1	0.83 ± 0.04	0.72 ± 0.04	0.81 ± 0.03	0.80 ± 0.03	0.78 ± 0.03	0.78 ± 0.07	NS	
C18:1	40.2 ± 0.62	38.4 ± 0.56	40.0 ± 0.60	39.9 ± 0.49	39.6 ± 0.65	40.0 ± 0.82	NS	
ΣMUFA	42.8 ± 0.68	40.8 ± 0.63	42.6 ± 0.61	42.8 ± 0.56	42.5 ± 0.70	42.7 ± 0.91	NS	
Polyunsaturated fatty acids (PUFA)								
C18:2n6	3.14 ± 0.24	3.52 ± 0.42	3.87 ± 0.23	3.62 ± 0.27	3.48 ± 0.18	3.81 ± 0.24	NS	
C18:3n3	0.77b ± 0.09	0.96ab ± 0.12	1.21a ± 0.12	1.04ab ± 0.11	1.04ab ± 0.07	1.14ab ± 0.11	0.05	
CLA	0.61b ± 0.12	0.87ab ± 0.16	0.99ab ± 0.11	1.15a ± 0.13	1.15a ± 0.09	1.24ab ± 0.19	0.008	
C20:3n6	0.09b ± 0.02	0.14ab ± 0.03	0.21a ± 0.03	0.14ab ± 0.02	0.17ab ± 0.02	0.18ab ± 0.05	0.01	
C20:3n3	0.21 ± 0.06	0.37 ± 0.11	0.43 ± 0.07	0.34 ± 0.07	0.32 ± 0.05	0.33 ± 0.08	NS	
C20:4n6	0.12 ± 0.04	0.19 ± 0.05	0.26 ± 0.04	0.19 ± 0.04	0.19 ± 0.03	0.22 ± 0.06	NS	
EPA	0.02 ± 0.00	0.02 ± 0.00	0.02 ± 0.01	0.02 ± 0.00	0.01 ± 0.00	0.01 ± 0.00	NS	
DPA	0.46 ± 0.07	0.33 ± 0.05	0.36 ± 0.04	0.41 ± 0.06	0.29 ± 0.03	0.26 ± 0.04	NS	
DHA	0.08 ± 0.05	0.04 ± 0.01	0.06 ± 0.01	0.02 ± 0.01	0.04 ± 0.01	0.11 ± 0.08	NS	
ΣPUFA	4.89 ± 0.43	5.56 ± 0.74	6.41 ± 0.52	5.76 ± 0.51	5.53 ± 0.35	6.06 ± 0.45	NS	
Unidentified Fatty Acids								
-	1.26 ± 0.17	0.98 ± 0.19	1.45 ± 0.11	1.63 ± 0.16	1.48 ± 0.19	1.78 ± 0.21	-	

Data are mean ± SEM. H= Highlander, MD=Merino Dohne, C= Corriedale, C PRO= Corriedale PRO, C x AM = Corriedale x Australian Merino, RM= Romney Marsh. For each fatty acid, mean values bearing different low case letters are significantly different. P= Significance level. NS= non-significant. i= iso, ai= anteiso, EPA= C20:5n3, DPA= C22:5n3, DHA= C22:6n3

Limited information was available in the scientific literature on the comparison of the fat content of CXAM and H. However, Jalloul et al. [13] report low lipid content in H lamb ranged between 1.07-1.18 g of lipids by 100 g of meat from *longissimus thoracis*. Nevertheless, lambs were housed in small pens and fed corn, citrus pulp, rice bran or soybean hulls. Thus, the difference in weight at slaughtering (30 kg versus 54 kg in our experiment), the kind of feed offered to animals, or both could explain the different fat content of meat. Regarding the other breeds studied in the present investigation, for C, the lipid content reported in meat obtained in conditions similar to Uruguay showed levels of around 3.65 g of lipids by 100 g of meat [14]. In the case of the present experiment, meat was from *longissimus thoracis*, while in the work of Lucas et al. [14], meat was from *longissimus lumborum*. In both experiments, animals were of similar age and fed pasture in the extensive production system. Another experiment in the extensive conditions of Uruguay compared C lamb produced on pasture, but at two different ages. Results showed lipids content of meat of 3.05 versus 5.92 g by 100 g of meat from *longissimus thoracis*, at 3-4 months and 12-13 months, respectively [15]. Compared to the reported in our experiment, the higher content of lipids could be explained by the nature of pasture more than by the difference of ages between the two works, that is, 12-13 months versus 11-12 months in our work. However, Diaz et al. [15] do not report the composition of pastures offered to the animals.

5.4.2. Fatty acids of glycerolipids

Regarding the fatty acids composition of glycerolipids, in the case of SAT, there are differences between the breeds for C16:0, C17:0 anteiso, C17:0 and C18:0 (TABLE II). C16:0 showed a higher content for MD compared to C. The fatty acid C16:0 (palmitic acid), among all SAT, is considered an atherogenic fatty acid and promotes inflammation [16]. Thus, its consumption is advised to be reduced but not avoided because of its important physiological role in lipids metabolism, particularly in neonates and infants [17]. Those authors state that both deficiency and excess palmitic acid in diet are detrimental to health. Probably, this concept could be valid for all ages. In the breeds studied in the present work, the level of C16:0 in meat ranged

21.1-26.1 g/100g fatty acids. These levels are of the same order of magnitude as those found in longissimus thoracis meat from lambs of different breeds produced in other countries. In fact, Cadavez et al [18], working on different local Iberian breeds produced in different rearing systems, i.e. extensive, semi-extensive and intensive, showed for C16:0 a value in g/100g of fatty acids ranging from 19.9-24.7. Diaz et al [15] compared meat from the longissimus dorsi, in its thoracic part, of different lamb breeds produced in the typical production systems of Spain, the United Kingdom, Germany and Uruguay. In that investigation, the composition for C16:0 expressed as g/100g fatty acids showed levels ranging 22.5-24.7. In the same investigation the breed evaluated in Uruguay was C, produced in two systems. One of them, called the heavy lamb, consisted of animals aged 12-13 months and the other, called the light lamb, consisted of animals aged 3-4 months. However, the palmitic acids levels were 24.66 % and 24.73 % in heavy lamb and light lambs, respectively. This apparent stability of contents of palmitic acid in meat, is probably because the intake in this fatty acid and the lipogenesis de novo for the same, act together to maintain a stable level of palmitic acid in tissues, even when the animals are of different ages as reported by Diaz et al. [15], for C lamb produced on pasture in Uruguay.

Another fatty acid, the anteiso C17:0ai is present in the meat of the six breeds (C17:0ai, TABLE II) and showed a level, expressed as g/100g fatty acids of all detected fatty acids, ranged from 0.35-0.58. However, only meat from MD presented a significantly lower level for this fatty acid in comparison to the other breeds. This fatty acid is mainly synthesized during the microbial fermentation processes in the rumen and, consequently, is typical of ruminant milk and meat products. Also, its amount of meat seems to be influenced by lamb fattening level when fed pasture [19]. However, in our work, although there is a difference between the breeds regarding their fattening, there is no clear relationship between lipids content in meat and level of anteiso C17:0 (TABLE II). The differences in the experimental conditions could explain the kind of muscles, breeds and pastures. There is scarce information about the nutritional importance in human health of anteiso C17:0. However, in the report of Vahmani et al. [20], the C17ai fails to present anti-carcinogenic properties in cultured MCF 7 cells, a mammary human cancer established cell line. In that investigation, the

iso C17:0 (C17:0i) presents a significant anti-carcinogenic effect in the same cell line MCF 7. Those promissory fatty acids and their potential effects on human health warrant future investigations to refine their action against some diseases [21]. In the same way, the odd fatty acids C17:0 presented values ranging from 1.35-1.67 when expressed as g/100g fatty acids (TABLE II). Breed C showed more C17:0 than the MD breed (TABLE II). These contents were higher than those reported by Diaz et al. [15] using lambs from the C breed produced on pasture in Uruguay but slaughtered at different ages. However, the rearing conditions were very similar to those of our experiments.

The combined impact of odd and branched fatty acids on consumer health, as detected in our experiment, will be discussed in section, titled ‘Monomethyl Branched and Odd Fatty Acids,’ of the present discussion.

Another fatty acid, the C18:0, showed a higher content for C than CxAM. The level for C18:0 observed in our experiment ranged from 18.3-21.1, being quantitatively the second saturated fatty acid present in the meat of lambs (TABLE II). The values of C18:0 reported in our work are of the same order as those reported by Lucas et al. [14] and Ramos et al. [22], even when crossbreed animals were used in those experiments. However, the work of Cadavez et al. [18], working on an Iberian local breed of lambs aged around 4-4.5 months and produced in different typical extensive, semi-extensive and intensive systems of Portugal and Spain, reported a level expressed in g/100g fatty acids ranged 12.3-15.4. This lower level of C18:0 reported in that investigation could be due to the age of the animals. But in another work comparing C lamb of 3-4 months with others aged 11-13 months showed levels of C18:0, in one part close to those observed in our work, and for another part, no difference has been detected between the animals of the two ages [15]. Therefore, it's crucial to consider the breeds used in these experiments, as they could be a significant factor contributing to the observed differences.

Concerning the human health effect when C18:0 is consumed, it has been defined that these fatty acids have a neutral fatty acid regarding cardiovascular diseases. However, some recent reports point to a potential adverse effect on human health. This controversial situation must be clarified in future investigations [16, 23].

In the case of MUFA, the C16:1 presented a significant difference between CPRO and CxAM versus MD. The levels of this fatty acid, expressed as g/100g fatty acids of all detected fatty acids, ranged from 1.57 to 2.10 (TABLE II). Those levels are of the same order as those reported in the experiment of Diaz et al. [15], Ramos et al. [22] and Lucas et al. [14] and close to or slightly lower than the levels observed by Cadavez et al., [18], even when different productive systems and breeds were used.

The fatty acid C16:1 has been suggested as a lipokine, particularly when synthesized endogenously. However, the positive health effects for humans consuming foods enriched with C16:1 are not clearly demonstrated, and the information remains controversial [24].

Regarding the PUFA, the level of C18:3n3 expressed as g/100g fatty acids ranged from 0.77-1.21, and C had more C18:3n3 than H (TABLE II). Compared with results from other reports, this fatty acid is present with levels of the same order as those reported by Ramos et al. [22] and slightly lower than values reported by Lucas et al. [14]. However, the levels of C18:3n3 in our work were lower than those reported by Diaz et al. [15], particularly when C is highlighted. Indeed, in that investigation, C, aged 3-4 months and others aged 11-14 months, present levels of C18:3n3 approximately almost three times those detected in the present work. This could be due to the quality of the pasture provided to the animals, as both experimental systems were extensive. As expected, in the work of Cadavez et al. [18], the animals reared in an extensive system present a higher content of C18:3n3 in comparison to those reared in a semi-extensive or an intensive system.

The fatty acid C18:3n3 (α -linolenic acid) is an essential fatty acid and precursor of other valuable fatty acids of the n-3 family. It is known to have a favourable effect on consumers' health, directly or after its conversion to C20:5n3 (EPA) and C22:6n3 (DHA). Those three fatty acids protect against cardiovascular diseases, cancer and probably some neurodegenerative diseases [16]. Thus, lamb meat in our experiment could be considered a good source of C18:3n3. Thus, lamb meat in our experiment could be considered a good source of C18:3n3. Unfortunately, the level of EPA and DHA detected in our experiment's lamb meat is not so high that it is not considered

relevant (TABLE II). The cause of those low levels of fatty acids will be investigated in future research.

Another fatty acid, CLA (Conjugated linoleic acid), showed levels expressed as g/100 g of fatty acids ranging from 0.61 to 1.24, and CPRO and CxAM presented higher than H (TABLE II). Those levels in CLA were in the same order as in the work of Diaz et al. [15], working with different breeds produced in Europe and Uruguay. In that experiment, only lamb from Spain showed a lower content of CLA compared to the other breeds used in the experiment. The levels of CLA observed in our work were also in accord with those reported by Ramos et al. [22], even when the animals were dietary supplemented with different protein levels. In the other part, the levels of CLA observed in the present work were also in accord with those reported by Lucas et al. [14].

CLA is a common fatty acid in significant quantities in ruminant meat and milk. Studies have linked CLA intake in humans to potential health benefits like anti-tumour effects, anti-obesity properties, and positive impacts on cardiovascular health [25].

5.4.3. Fatty acids of glycerophospholipids

For SAT, the fatty acid C17:0 showed levels expressed as g/100g fatty acids ranging from 0.95-1.31. The breed H presented a higher content than RM for this fatty acid (TABLE III).

TABLE III

Fatty acids composition (g/100 g fatty acids) of glycerophospholipids present in Longissimus thoracis muscle from lambs of different breeds produced on pasture.

Breeds							
	H (n=15)	MD (n=11)	C (n=11)	CPRO (n=15)	CxAM (n=15)	RM (n=4)	P≤
Saturated Fatty Acids (SAT)							
C16:0	22.5 ±1.38	20.1 ±1.51	18.8 ±1.45	19.5 ± 1.49	18.6 ±1.09	17.4 ±1.61	NS
C17:0	1.31a ±0.08	1.19ab ±0.10	0.98ab ±0.09	1.08ab ±0.09	1.00ab ±0.06	0.95b ±0.24	0.05
C18:0	18.8 ±0.57	17.7 ±0.76	18.0 ±0.47	17.6 ±0.47	17.1 ±0.43	18.0 ±2.27	NS
C22:0	0.18 ±0.04	0.31 ±0.08	0.33 ±0.05	0.33 ±0.06	0.33 ±0.04	0.40 ±0.05	NS
Σ SAT	42.8 ±1.72	39.3 ±2.09	38.1 ±1.77	38.6 ±1.82	37.0 ±1.46	39.3 ±2.09	NS
Monounsaturated Fatty Acids (MUFA)							
C16:1	1.21 ±0.06	1.17 ±0.14	0.82 ±0.08	1.06 ±0.14	1.03 ±0.06	0.87 ±0.14	NS
C17:1	0.89ab ±0.08	0.81ab ±0.07	0.74ab ±0.06	0.71b ±0.06	0.92b ±0.08	1.30a ±0.20	0.005
C18:1	37.2a ±0.97	34.5ab ±1.35	31.7ab ±1.19	32.3b ±1.28	32.4b ±0.96	31.9ab ±1.30	0.008
ΣMUFA	39.3a ±1.00	36.5ab ±1.25	33.3b ±1.25	34.1ab ±1.41	34.6ab ±0.96	36.5ab ±1.26	0.029

	Breeds						
	H (n=15)	MD (n=11)	C (n=11)	C PRO (n=15)	CxAM (n=15)	RM (n=4)	P≤
Polyunsaturated Fatty Acids (PUFA)							
C18:2n6	8.92b ±0.87	12.2ab ±1.35	13.3ab ±1.01	12.8ab ±1.32	13.3a ±0.78	13.5ab ±1.13	0.02
C18:3n3	2.29b ±0.45	3.48ab ±0.56	4.71a ±0.59	3.92ab ±0.56	4.12ab ±0.41	4.46ab ±0.66	0.02
C20:3n6	0.36b ±0.07	0.68ab ±0.14	0.85a ±0.11	0.74ab ±0.11	0.70ab ±0.08	0.76ab ±0.16	0.01
C20:3n3	1.44 ±0.41	2.40 ±0.51	3.11 ±0.47	2.87 ±0.51	2.83 ±0.37	3.17 ±0.67	NS
C20:4n6	1.29 ±0.48	1.64 ±0.41	2.66 ±0.55	2.34 ±0.54	2.54 ±0.46	3.15 ±0.91	NS
EPA n3	0.30 ±0.11	0.22 ±0.09	0.34 ±0.10	0.29 ±0.08	0.35 ±0.09	0.45 ±0.20	NS
DPA	0.67 ±0.21	1.01 ±0.25	1.57 ±0.26	1.43 ±0.26	1.58 ±0.32	1.64 ±0.40	NS
DHAn3	0.31 ±0.04	0.27 ±0.04	0.35 ±0.05	0.26 ±0.02	0.26 ±0.02	0.31 ±0.06	NS
Σ PUFA	15.6 ±2.48	21.9 ±3.16	26.9 ±2.88	24.6 ±3.20	25.7 ±2.27	21.9 ±3.16	NS
Unidentified Fatty Acids							
-	2.40 ±0.26	2.34 ± 0.32	1.78 ± 0.45	2.66 ± 0.33	2.92 ± 0.27	1.66 ± 0.80	-

Data are mean ±SEM. H= Highlander, MD=Merino Dohne, C= Corriedale, C PRO= Corriedale PRO, C x AM = Corriedale x Australian Merino, RM=Romney Marsh. For each fatty acid, mean values bearing different low case letters are significantly different. P= Significance level. NS= non-significant. EPA= C20:5n3, DPA= C22:5n3, DHA= C22:6n3.

There are no differences in total SAT for the breeds used in our work (TABLE III). For the other part, the comparison of our results to other reports showed that the level of C16:0 was in the same order as the results reported by Aurousseau et al. [26] and slightly and slightly higher compared to the results reported by Popova [27], in both experiments, animals were fed pasture. However, our results for this fatty acid were higher than those reported by Garcia et al. [28]. In this last work, using animals from the Merino breed, which is typical for wool production, could explain those differences. Furthermore, the lambs were fed shrub grass steppes, which could explain this result. For the C18:0, the levels expressed as g/100g fatty acids reported by Popova [27] were of the same order as those observed in our work. However, the level of C18:0 reported by Aurousseau et al. [26] and Garcia et al. [28] were lower when compared to our results for this fatty acid (TABLE III). As expressed before, differences due to the breed pasturage, or both, could explain those results.

For MUFA, C17:1 presented a level expressed as g/ 100 g fatty acids ranging from 0.71-1.30. RM presented a higher level than CPRO and CxAM (TABLE III). There are scarce results in the scientific literature on the content of this fatty acid in lamb meat. However, compared to our results, Garcia et al. [28] reported approximately three times more C17:1, expressed in g/100g fatty acids.

In the case of C18:1, the levels expressed as g/100g fatty acids ranged from 31.7-37.2, and H presented a higher level than CPRO and CxAM (TABLE III). Those levels are of the same order as those reported by Aurousseau et al. [26] and Popova [27], much higher compared to the results of Garcia et al. [28]. The same explanation proposed before to explain the differences between our results and those of Garcia et al. [28] could be proposed again here. That is, lambs were Merino, a typical breed for wool production, fed shrub grass steppes.

For PUFA, the fatty acid C18:2n6 presented a level expressed as g/100 g of fatty acids ranging from 8.92 to 13.5 (TABLE III). The C exhibit more C18:3n3 than H (TABLE III). This range of levels was of the same order as the levels reported by Aurousseau et al. [26] and Garcia et al. [28] but lower than those reported by Popova [27]. In our work, CxAM exhibits more C18:2n6 in comparison to H. This fatty acid is the most represented in tissue glycerophospholipids and is the essential precursor of

other fatty acids of the n-6 family, for example, the C20:4n6 (arachidonic acid). At the same time, this last is a prevalent precursor of many pro-inflammatory eicosanoids, leukotrienes, and thromboxanes, among other biomarkers of inflammation [16].

In our study, the levels of C18:3n3 ranged from 2.29 to 4.71 g/100 g of fatty acids. These levels were found to be similar to or slightly higher than those reported in other studies [26, 27, 28]. It's worth noting that in those experiments, the animals were fed pasture or shrub grass steppes. [28]. The favourable effect of these fatty acids on consumers' health is presented in the "Fatty acids of glycerolipids" section above. Another fatty acid that showed differences between the breeds used in our work was the 20:3n6 (TABLE III). The same levels expressed as g/ 100 g of fatty acids ranged from 0.36-0.85, and C showed more 20:3n6 than H (TABLE III). The values detected in our work were in the same order as those reported by Popova [27] and Garcia et al. [28] with lamb-fed pasture. This fatty acid has been implicated, not alone but concomitantly with other n-6 fatty acids, in the prevalence of higher severity of depressive and anxiety symptoms in patients with depression [3]. Furthermore, other pathologies linked to the inflammatory process could relate to this fatty acid's metabolism [29]. Considering the levels of these fatty acids in lamb meat and their effect on human health, as described above, it could be exciting and vital to consider them as relevant fatty acids to be considered in future investigations on lamb meat.

Finally, although there is no difference between the breeds for glycerophospholipids for C20:4n6, EPA, DPA and DHA, the Authors of the present work think it justified to compare the levels of those fatty acids obtained in our work to levels reported elsewhere. Indeed, considering the nutritional importance of these fatty acids for human health [30, 31], it will be interesting to evaluate the contents of those fatty acids in the meat of lamb produced on pasture in Uruguay. For C20:4n6, the levels recorded in the work expressed as g/100 g fatty acids ranged from 1.29-3.15 (TABLE III). Those values were lower than those reported by Aurousseau et al. [26] and Garcia et al. [28]. In the case of the work of Popova [27], the value reported was substantially more elevated compared to our work and those cited before, that is, 8.23% % and 7.21 % for muscles longissimus lumborum and semimembranosus, respectively. Consumption by humans of elevated amounts of C20:4n6 is not advised

because this fatty acid is a precursor of prostanoids of series 2, leukotrienes of series 4, and many other eicosanoids, all of them promoting inflammation and causing vasodilatation. This fatty acid also could elevate the risk of hypertension and arteriosclerosis [16, 32].

For EPA, the levels detected in our work ranged from 0.22-0.45 g/100g (TABLE III), while the values reported by Aurousseau et al. [26] were 4.1 and 8.23 for Popova [27], both results expressed as g/100 g fatty acids. However, in the work of Garcia et al. [28], the level of EPA was 1.60, described as g/100 g fatty acids. For DPA, Aurousseau et al. [26] and Garcia et al. [28] reported levels of 1.10 and 1.35 as g/100 g of fatty acids, respectively. In the case of Popova [27], the reported content in DPA was 3.28 for longissimus lumborum and 2.76 for semitendinosus; both expressed as g/100 g of fatty acids. In the current work, the observed levels ranged from 0.26-0.35 for the DHA, expressed as g/100 g of fatty acids. In comparison with the report of Popova [27], the levels were 0.74 and 0.57 as g/100 g of fatty acids for longissimus lumborum and semitemdinosus, respectively. In the case of Garcia et al. [28], the reported levels were 0.56. In the work of Aurousseau et al. [26], the amount of DHA in lamb's meat should have been reported.

The levels of those valubles n-3 PUFA in glycerophospholipids, that is, EPA, DPA, and DHA, as detected in our work, clearly present a lower content compared to other reports previously cited. Those differences could be explained, on the one hand, by the environmental condition of the rearing and the global management of the animals, which include the kind and the quality of offered pastures [33]. On the other hand, the breed used in our work could have a reduced capacity to efficiently convert the C18:3n3 to EPA, DPA and DHA, as stated by Sinclair et al. [34]. In this sense, it could be noted that the levels of C18:3n3, the precursor of EPA, DPA and DHA, present in our work at a higher level in meat in comparison to those levels reported by Aurousseau et al. [26], Popova [27] and Garcia et al. [28]. That means there was more storage of this fatty acid in the muscle rather than being converted into other n-3 fatty acids [34]. This hypothesis needs to be verified in future investigations in lamb.

5.4.4. Monomethyl branched and odd fatty acids

In TABLE IV, the total of monomethyl branched-chain fatty acids (BCFA) detected in our study.

The total BCFA levels ranged from 1.23-1.57g/100g. There are no differences between the breeds studied in our work. That range of values was slightly higher than those reported by Gomez-Cortes et al. [35] and Pena-Bermudez et al. [36] but somewhat lower than those presented (control group) by Mele et al. [37]. The animals in all three experiments were given concentrate, and the outcomes were measured as g/100 g of fatty acids. The differences in breed, age, feeding system and the kind of management between the works determined differences in the reported levels of BCFA. The review by Vahmani et al. [38] demonstrates the varying levels of BCFA in ovine meat.

However, the food offered, the composition of concentrate, or the botanical composition of pasture could be the most important factor explaining the differences in BCFA in lamb meat. For example, C in the present experiment presented a level of BCFA of 1.43 g/100 g fatty acids which included C15:0 iso and anteiso, C16:0 iso and C17:0 iso and anteiso (TABLE II). In another experiment by our laboratory using C, meat presented a level of BCFA of 0.21g/100g fatty acids and only C15:0 iso and anteiso were detected. No other BCFA were detected above the 0.01 g/ 100 g fatty acids [14]. In both experiments, the animals were of the same age (11-12 months), reared extensively and fed pasture, and slaughtered in similar commercial conditions, and all the procedures for extraction and detection of the fatty acids were identical to those described in the present work. There are only two differences between the two works. One of them was the muscle evaluated, longissimus thoracis, in the present work versus the longissimus lumborum in the work of Lucas et al. [14]. The other was the type of offered pastures regarding their botanical composition, mainly oats and legumes in the present work (see TABLE I) versus grasses in the work of Lucas et al. [14]. Some reports support the concept that the kind, composition, and type of pasture could influence the level and type of fatty acids, including BCFA, in lamb meat [39].

TABLE IV

Branched and odd fatty acids (g/100g) of meat from Longissimus thoracis muscle of lambs of different breeds produced on pasture

	Breeds						P≤
	H (n=15)	MD (n=11)	C (n=11)	C PRO (n=15)	CxAM (n=15)	RM (n=4)	
BCFA	1.39± 0.07	1.23± 0.09	1.43± 0.08	1.46± 0.06	1.42± 0.06	1.57± 0.14	NS
BCFA i	0.76± 0.05	0.71± 0.05	0.78± 0.04	0.80± 0.03	0.76± 0.04	0.85± 0.10	NS
BCFA ai	0.63ab± 0.03	0.52b± 0.05	0.65ab± 0.04	0.66ab± 0.03	0.67ab± 0.02	0.73a± 0.05	0.03
Odd Fatty Acids	4.23± 0.15	3.89± 0.18	4.41± 0.17	4.39± 0.11	4.18± 0.13	4.38± 0.38	NS

Data are mean ±SEM. H= Highlander, MD=Merino Dohne, C= Corriedale, CPRO= Corriedale PRO, CxAM = Corriedale x Australian Merino, RM= Romney Marsh. Within lines, mean values bearing different low case letters are significantly different. P= Significance level. NS= non-significant. BCFA= sum of total branched fatty acids, i= iso, ai= anteiso, BCFAi= sum C15:0i + C16:0i + C17:0i, BCFAai= sum C15:0ai + C17:0ai, Odd Fatty acids= C15:0 + C15:0i + C15:ai + C17:0 + C17:0i + C17:0ai + C17:1. All calculations were performed on base of results of TABLE II

Indeed, in the rumen, the interrelation between the microbial populations [40], the specific fatty acids synthetases and the composition of pasture regarding fatty acids and amino acids will lead to different kinds of fatty acids present in ruminant meat [41, 42]. In the case of amino acids, the content of leucine, isoleucine and valine in the pasture seems to influence the kind of final BCFA present in ruminant meat [41]. This effect of amino acids in the composition of meat regarding the BCFA could partially explain the differences observed between the present experiment and that of Lucas et al. [14]. Indeed, the differences between the two investigations could account for the richness of legumes and oats in those three amino acids, in comparison to the work of Lucas et al. [14], where the animal was fed quantitatively mainly gramineae [43, 44]. This interesting point needs more exploration and investigation to understand better how BCFA, particularly those linked to positive effects on health, could be incorporated in lamb meat, thanks to different kinds of offered pastures.

Regarding the BCFA iso (BCFAi) and anteiso (BCFAai), the level of the former ranged from 0.71-0.85, expressed as g/100 g fatty acids (TABLE IV). There were no differences between the breeds studied in this work. The range of BCFAi observed in our work was of the same order as those reported by Gomez-Cortes et al. [35] and Mele et al. [37] but higher than the values reported by Pena-Bermudez et al. [36]. Note that in those three reports, the lambs were fed concentrate. BCFAai's range was between 0.52-0.73 g /100 g of fatty acids, and RM presented a higher content than MD (TABLE IV). The range of BCFAai observed here is of the same order as those reported by Mele et al. [37] but higher than those reported by Gomez-Cortes et al. [35] and Pena-Bermudez et al. [36]. Indeed, the explanation presented before that in the rumen, the interrelation between the microbial populations, the specific fatty acids synthetases and the composition of pasture regarding fatty acids and amino acids leading to the different kinds of BCFA in meat could be introduced newly here. This is particularly true for BCFAi and BCFAai regarding the amino acid composition of pasture [41].

In the case of the Odd fatty acids detected in our work, the range of values was 3.89-4.41. There were no differences between the breeds studied in our work (TABLE IV). That range was in the same order of levels reported by Mele et al. [37], slightly

higher than the values of Garcia et al. [28], but markedly higher in comparison to values reported by Gomez-Cortes et al. [35] and Pena-Bermudez et al. [36]. As indicated before, the lambs used in the works of Mele et al. [37], Gomez-Cortes et al. [35] and Pena-Bermudez et al. [36] were fed concentrated, while the lambs in the work of Garcia et al. [28] were fed shrub grass steppes. As for BCFA, the Odd fatty acids are influenced by the rumen metabolism, which is, in turn, influenced by its microbial population, the primers present for lipogenesis, that is, the balance of acetyl-CoA versus propionyl-CoA, and, of course, the kind of food consumed by the animals [41]. This could explain the observed differences between the different studies highlighted here. In the same direction, Lucas et al. [14] reported level of Odd fatty acids of 2.40 g/ 100 g fatty acids for C, a lower level in comparison to the value observed in the present work (4.41 g/ 100 g fatty acids) for the same breed C (TABLE IV). As stated before, the main difference between the two studies was the botanical type of pasture offered to the lamb, if the muscle difference that is longissimus thoracis here versus longissimus lumborum in Lucas et al. [14], is ruled-out as main factor, to explain the differences.

As stated, before in the text, individual BCFA and odd fatty acids have some beneficial effects on consumers' health related to those fatty acids [21]. More investigation must be undertaken to improve knowledge about the impact of this kind of component present in the meat and milk of ruminants on human health.

5.4.5. Lipids health indices

In TABLE V, some indices were grouped to help determine the nutritional characteristics associated with the health of consumers of this kind of lamb meat. The sum of n-6 fatty acids presented a range between 3.35-4.34 g/ 100 g of fatty acids.

TABLE V Lipids health indices, of meat from longissimus thoracis muscle of lambs of different breeds produced on pasture

Data are mean \pm SEM. H= Highlander, MD=Merino Dohne, C= Corriedale, CPRO= Corriedale PRO, C x AM = Corriedale x

	Breeds						P≤
	H (n=15)	MD (n=11)	C (n=11)	CPRO (n=15)	CxAM (n=15)	RM (n=4)	
$\Sigma n-6$	3.35 \pm 0.29	3.85 \pm 0.49	4.34 \pm 0.29	3.94 \pm 0.32	3.83 \pm 0.23	4.21 \pm 0.27	NS
$\Sigma n-3$	1.53 \pm 0.14	1.72 \pm 0.25	2.08 \pm 0.23	1.82 \pm 0.19	1.70 \pm 0.13	1.85 \pm 0.19	NS
n-6/n-3	2.25 \pm 0.12	2.32 \pm 0.08	2.24 \pm 0.14	2.25 \pm 0.09	2.31 \pm 0.06	2.31 \pm 0.13	NS
P/S	0.10 \pm 0.01	0.11 \pm 0.02	0.14 \pm 0.01	0.12 \pm 0.01	0.12 \pm 0.01	0.13 \pm 0.01	NS
AI	0.73 \pm 0.06	0.89 \pm 0.09	0.64 \pm 0.04	0.73 \pm 0.06	0.78 \pm 0.05	0.63 \pm 0.05	NS
TI	1.70 \pm 0.08	1.80 \pm 0.14	1.51 \pm 0.07	1.55 \pm 0.07	1.62 \pm 0.07	1.51 \pm 0.05	NS
h/H	1.87ab \pm 0.11	1.64b \pm 0.13	2.12a \pm 0.11	1.93ab \pm 0.09	1.80ab \pm 0.09	2.08ab \pm 0.09	0.05

Australian Merino, RM= Romney Marsh. Within lines, mean values bearing different low case letters are significantly different. P= Significance level. NS= non-significant. $\Sigma n-6$ = total n-6 fatty acids, $\Sigma n-3$ = total n-3 fatty acids, EPA= C20:5n3, DHA= C22:6n3, P/S= PUFA/SAT ratio, AI= atherogenic indices, TI=thrombogenic indices, h/H=hypocholesterolemic indices, BCFA= total branched fatty acids, i= iso, ai= anteiso, Odd FA= odd fatty acids

These levels are the same order as those reported by Lucas et al. [14], working on C, and Ramos et al. [22] using crossing between C and MD; both investigations were conducted in the condition of Uruguay. However, another investigation also using C [15] reported slightly higher values in n-6 fatty acids, 5.14 versus 4.34 g/ 100 g fatty acids / 100 g, as reported in this work (TABLE V). Note that the animals were of similar age in those experiments, reared extensively and fed pasture. Outside of Uruguay, using local breed reared extensively on pasture, Elaffifi et al. [45] reported 3.59 g of n-6 / 100g of fatty acids. A value within the range observed in our work. However, other trials using other breeds fed pastures showed higher levels of n-6 fatty acid, such as in the work of Faria et al. [46] and Garcia et al. [28], 10.56 and 9.23 g /100 g fatty acids, respectively. This last work used Merino lamb-fed shrub grass steppes.

Regarding the n-3 fatty acids, the same pattern as reported for the n-6 has been noted. Indeed, the works of Lucas et al. [14], Ramos et al. [22] and Elaffifi et al. [45] reported values within the range observed in the present work, while the reports of Diaz et al. [15], Garcia et al. [28] and Faria et al. [46] reported a much higher level of n-3 fatty acids in meat.

In the case of the n-6:n-3 ratio, the range observed in our work was between 2,24-2.31. These values were in the same order as those reported by Cadavez et al. [18], Ramos et al. [22], Garcia et al. [28] and Elaffifi et al. [45]. In contrast, the values observed in our work were slightly lower compared to the report of Faria et al. [46] or lower, particularly for C, when reared in similar productive conditions in Uruguay [15]. In practice, the content of n-6 and n-3 and their ratio in the meat of the animals evaluated in the current work seem to have values in accord with other results reported in lamb. Albeit there is no specific advice about adequate n-6 and n-3 fatty acids regarding human health, the ratio between those two fatty acids has been recommended to be near 4-5 [47], or even between 1:1 and 2:1 [48]. The meat of the animals evaluated in the present work presents a ratio of n-6:n-3 in accord with the recommendation of 2:1. However, nowadays, that ratio becomes open to question about its usefulness regarding human health [49]. Future investigations should

improve the knowledge to establish better parameters related to the human consumption of lipids and the fatty acids in lamb meat.

Another index established as a parameter related to human health was the ratio between PUFA and SAT (P/S). In our experiment, the P/S ratio observed varied between 0.10 and 0.14 (TABLE V). Those values were of the same order as those reported by Ramos et al. [22] but largely below those reported by Diaz et al. [15], Cadavez et al. [18], Garcia et al. [28], and Faria et al. [46]. The recommended levels of P/S in different kinds of meat to ensure adequate health in humans regarding cardiovascular diseases must be between 0.4 and 1 [50]. Thus, the meat of the animals used in our work is below the recommended level, as reported in TABLE V. Therefore, this point justifies particular attention in future investigations.

The other indices used in our work to assess the potential protection against cardiovascular disease were AI, TI and h/H [51]. For AI, the value observed in our work ranged from 0.63-0.89 (TABLE V). The advised level of AI must be as low as possible. The range observed was of the same order or slightly higher than those reported by Cadavez et al. [18], Belhadj et al. [52] and the compilation work by Procisur-IIICA [53]. For TI, the values observed in our work ranged from 1.51-1.80 (TABLE V). Those levels are higher than those reported by Cadavez et al. [18], Belhadj et al. [52] and the compilation of Procisur-IIICA [53]. The recommended value of TI in meat must be as low as possible to reduce the thrombogenic effects in humans [54].

Regarding h/H indices, the values observed in the current work ranged from 1.64-2.12, and the C showed a higher index than MD (TABLE V). For that index, the recommended value of meat must be as high as possible to minimize the risk of hypercholesterolemia leading to cardiovascular diseases [55]. The range of levels observed in our work was of the same order or slightly lower than those reported by Belhadj et al. [52], working with four local breeds fed pasture in Morocco, and Murariu et al. [56] in Romania using Karakul lamb-fed pasture and supplemented with hay and cereals in the winter season.

Some of the presented indices, such as P/S and TI, were not within the recommended value; therefore, more investigation must be undertaken to improve

those parameters. This could be done through the modification of the feeding system of the animals using different kinds of pasture. This point is an essential challenge for ovine production in Uruguay as a way to help farmers promote their products based on consumers' health. Some of the presented indices, such as P/S and TI, were not within the recommended value; therefore, more investigation must be undertaken to improve those parameters. This could be done through the modification of the feeding system of the animals using different kinds of pasture. This point is an essential challenge for ovine production in Uruguay as a way to help farmers promote their products based on consumers' health.

5.4.6. Enzyme activity indices

Enzyme indices for desaturases Δ-9, Δ-5 and Δ-6, elongase, and thioesterase have been calculated in an attempt to detect differences in the lipids metabolism between the breeds studied in the present work. These indices are generally used as surrogates to measure true enzyme activities. This procedure has also been used in the medical field as a simple way to evaluate the activities of enzymes such as desaturases, elongases and thioesterases in some human pathologies [57].

The enzyme activities were presented in TABLE VI, and it can be seen that C, CPRO and CxAM have a much more active Δ-9-C16 than MD.

TABLE VI

Enzymes indexes of fatty acid metabolism estimated on the basis of fatty acid composition of Longissims thoracis muscle of lambs of different breeds produced on pasture

	Breeds						P≤0.05
	H (n=15)	MD (n=11)	C (n=11)	C PRO (n=15)	CxAM (n=15)	RM (n=4)	
Δ-9 - C16	6.98ab ± 0.46	5.88b ± 0.56	7.91a ± 0.44	8.22a ± 0.27	7.97a ± 0.40	8.11ab ± 0.47	0.02
Δ-9 - C18	66.4 ± 0.89	67.0 ± 0.48	65.5 ± 0.77	68.0 ± 0.81	68.5 ± 0.90	65.8 ± 1.38	NS
Δ-9- C16+C18	48.6 ± 0.87	47.1 ± 1.02	49.7 ± 0.75	50.1 ± 0.53	49.4 ± 0.86	50.0 ± 1.00	NS
Δ-5	45.0 ± 5.10	49.7 ± 6.60	51.6 ± 3.92	46.9 ± 4.85	50.4 ± 2.98	53.5 ± 5.03	NS
Δ-6	2.51b ± 0.35	3.49ab ± 0.47	4.83a ± 0.51	3.51ab ± 0.38	4.33ab ± 0.42	4.62ab ± 1.05	0.005
Elongase	0.88ab ± 0.06	0.75b ± 0.05	1.01a ± 0.05	0.85ab ± 0.05	0.77b ± 0.04	0.99ab ± 0.07	0.008
Thioesterase	10.1 ± 0.55	7.99 ± 0.51	9.08 ± 0.47	8.26 ± 0.51	8.46 ± 0.52	9.38 ± 1.35	NS

Data are mean ±SEM. H= Highlander, MD=Merino Dohne, C=Corriedale, CPRO=Corriedale PRO, C x AM = Corriedale x Australian Merino, RM= Romney Marsh. Within lines, mean values bearing different low case letters are significantly different. P= Significance level, NS= non-significant, C16= palmitic acid, C18= stearic acid. Δ-9= Δ-9-desaturase, Δ-5= Δ-5-desaturase, Δ-6= Δ-6-desaturase, indexes of Δ-9, Δ-5 and Δ-6 desaturases, and elongase (ratio C18:0/C16:6) and thioesterase (ratio C16:0/C14:0) indexes were calculated according to del Puerto et al. [11]

This result could explain the differences in C16:1 content, which was reported for glycerolipids in TABLE II, for CPRO and CxAM, but not for C.

In the case of the enzyme Δ -9-C18 and the sum of the activities of both enzymes Δ -9-C16+C18, there are no differences between the animals studied in the present work (TABLE VI). Δ -9 enzyme introduces a cis- double bond in the D9 position between carbons 9 and 10, and the preferred substrates are palmitoyl-CoA for Δ -9-C16 and stearoyl-CoA for Δ -9-C18, which lead to their conversion into palmitoleyl-CoA and oleoyl-CoA, respectively [58]. MUFA, particularly C18:1, are important for membrane structure and function based on phosphoglycerides [59]. However, globally, there is no clear difference between the breeds regarding the de novo synthesis and the deposition of MUFA in glycerolipids and glycerophospholipids in the longissimus thoracis muscle. The fact that the animals have been reared in identical conditions, that is, fed the same pasture and management, could have minimised the possible enzyme expression differences between the breeds.

The enzyme indices for Δ -5 do not show either difference between the animals, while for Δ -6, C has a higher activity than H (TABLE VI). This result probably explains the higher content in C18:3n3 for C compared to H, as well as for glycerolipids than for glycerophospholipids (TABLES II and III). Indeed, Δ -5 and Δ -6 desaturases are crucial for the synthesis of PUFA [46, 57].

In the case of the elongase activities, C showed higher activity compared to MD (TABLE VI). The enzymes elongase add two carbon atoms to the fatty acid C16:0, obtaining the fatty acid C18:0, but also elongate other fatty acids from the two essential fatty acids C18:2n6 and C18:3n3 [60, 61]. However, the comparison between C and MD regarding the content of C18:0 and PUFA does not show differences for either glycerolipids or glycerophospholipids (TABLES II and III). The differential activity observed for elongases between C and MD was probably too small to significantly affect the content of C18:0 and PUFA in the meat of both breeds. As stated before, for desaturases, the fact that the animals have been reared in identical conditions, fed the same pasture, and conducted within the same extensive system could possibly have minimised the differences between breeds for elongase activities.

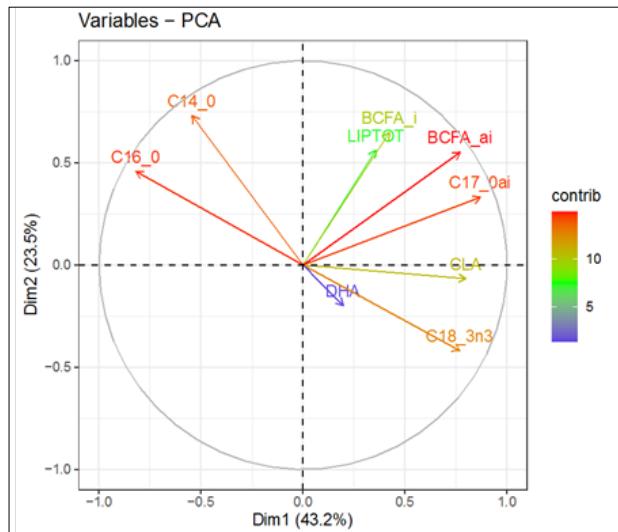
For the thioesterase indices, there are no differences between the six breeds studied here (TABLE VI). Thioesterase is part of the fatty acid synthase enzyme complex encoded by the FASN gene in mammals. It principally regulates the formation of C16:0 as the final product and C14:0 as a minor one [62]. Taking into account, as reported above, the implication of C16:0 in cardiovascular diseases in humans, hence it could be interesting to investigate how the thioesterase activity in lamb meat can be modulated. That focus could help to improve the nutritional and health quality of meat regarding the content of C16:0 in meat. Future research could open the way in that direction.

5.4.7. Interrelations among fatty acids of glycerolipids and glycerophospholipids

A principal component analysis carried out on total lipids and selected fatty acids of glycerolipids of lamb meat (FIG. 1a) shows that the two first principal components accounted for 66.7 % of the data variability. The first component (43.2 %) was positively correlated with C17:0ai ($r=0.870$), 18:3n3 ($r=0.769$), CLA ($r = 0.798$) and BCFAai ($r= 0.771$), and associated negatively to C14:0 ($r=0.544$) and C16:0 ($r=0.820$).

The second component that explains 23.5% of the data variability was positively associated mainly with intramuscular fat content ($r=0.562$), C14:0 ($r=0.731$) and BCFAi ($r=0.648$). The principal component analysis shows that lamb meat with a higher content of linolenic acid tends to have a lower content of saturated fatty acids, C14:0 and C16:0. When the individual observations for glycerolipids are projected in the two-dimensional space (FIG. 1b), there is evidence that H and MD genotypes are differentiated from others by CLA, BCFAai, C17:0ai, and 18:n3.

a)



b)

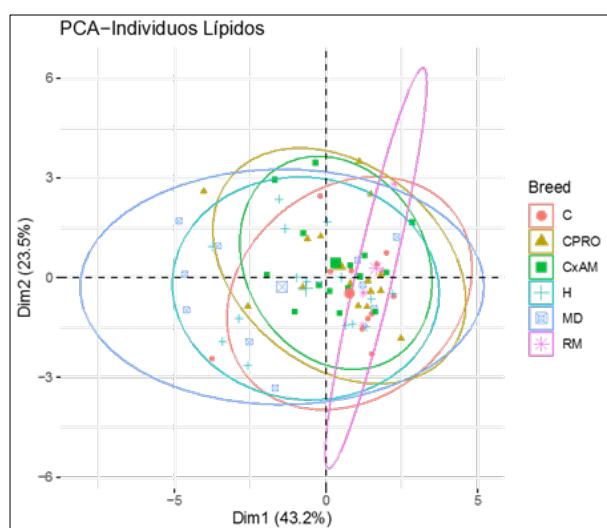
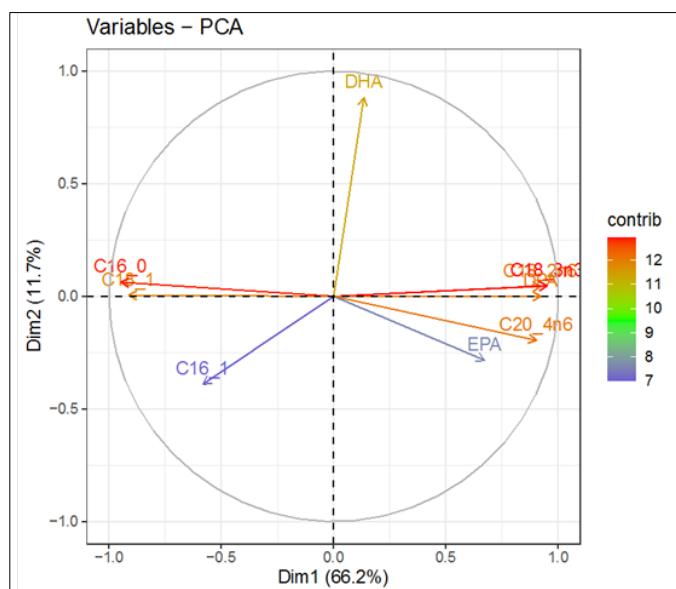


FIGURE 1. a) Variables factor map of compositional lipid metabolism of glycerolipids related to human health of lamb meat from six breeds. Liptot= total lipids; C14:0= myristic acid; C16:0= palmitic acid; C17:0ai=margaric acid anteiso; BCFAai= anteiso branched chain fatty acid; BCFAi=iso branched chain fatty acid; C18:3n-3=linolenic acid; CLA= conjugated linoleic acid; DHA= C22:6n3. b) Individuals factor map of compositional lipid metabolism of glycerolipids in lamb meat grouped by breed, with ellipses superimposed at $\alpha = 0.95$. C=Corriedale; CPRO=Corriedale Pro; CxAM=Corriedale x Australian Merino; H=Highlander; MD=Merino Dohne; RM= Romney Marsh

On the other side, the variability of RM is associated with component two, BCFAi, intramuscular fat content and C14:0. In terms of lipid attributes selected, lamb meat of C, CPRO, and CxAM overlap in quality attributes, associated with higher CLA and 18:3n3 and H with the lower content and also lower content of lipids. RM shows a higher level of BCFAai, possibly related to intramuscular fat content. When a principal component analysis was carried out for glycerophospholipids, a clearer association among variables was observed (FIGS. 2a and 2b). Two components explain the 77.9% of the total variability.

a)



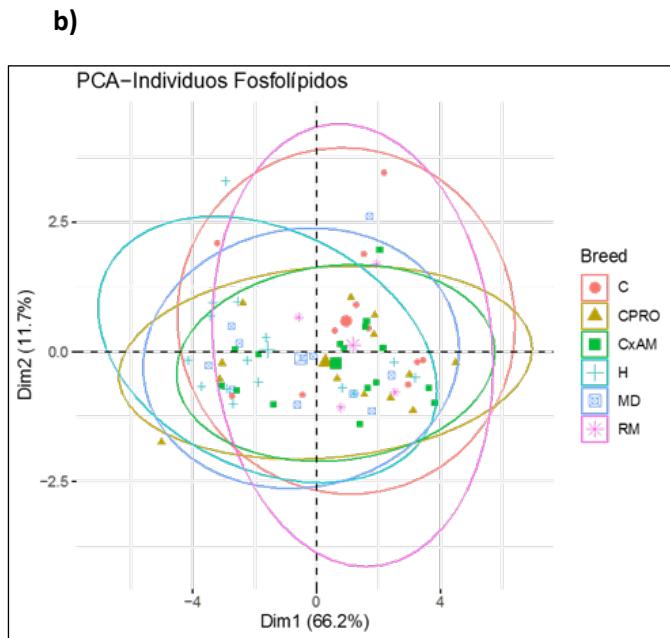


FIGURE 2. a) Variables factor map of compositional lipid metabolism of glycerophospholipids fraction related to membrane structure of lamb meat from six breeds. C16:0= palmitic acid; C16:1= palmitoleic acid; C18:1= oleic acid; C18:2n6= linoleic acid; C18:3n-3=linolenic acid; C20:4n6= arachidonic acid; EPA= C20:5n3; DPA=C22:5n3; DHA= C22:6n3. b) Individuals factor map of compositional lipid metabolism associated to glycerophospholipids fraction of lamb meat grouped by breed, with ellipses superimposed at $\alpha = 0.95$. C=Corriedale; CPRO=Corriedale Pro; CxAM=Corriedale x Australian Merino; H=Highlander; MD=Merino Dohne; RM=Romney Marsh

The first component that explains the 66.2% of the variability is positively and hardly associated with C18:2n6 ($r=0.922$), C18:3n3 ($r=0.951$), C20:4n6 ($r=0.903$) and DPA ($r=0.922$) and negatively with C16:0 ($r=0.948$) and C18:1 ($r=0.913$). Component two, which explains 11.7% of the variability, is mainly associated positively with DHA ($r=0.882$). Concerning individual observations for glycerophospholipids, only CPRO is related to the variables of the first component. In contrast, for individuals of C, a relation is evidenced for the variables that affect component two, the DHA.

5.5. Conclusions

The results of the study show overlapping among breeds related to compositional lipid metabolism, except for a few relevant fatty acids such as C16:0, C18:3n3 and CLA for glycerolipids, and C18:1, C18:2n6 and C18:3n3 for glycerophospholipids. Likewise, other differences were outlined for BCFAai, h/H and enzyme activity of Δ-9-C16, Δ-6 and elongase. However, the differences are between two or three breeds of the six studied in the present investigation and not for all those relevant fatty acids.

Thus, it can be said that overall, the studied breeds present good lipid nutritional indicators in comparison with the results of other research in lambs, except for EPA and DHA fatty acids, as those breeds present a relatively low content in contrast to the values indicated in some reports from the scientific literature. This last point will be considered in our future studies to improve the final composition of meat of those breeds, with the most relevant n-3 fatty acids regarding consumers' health.

As mentioned throughout the text, the animals were fed and managed in identical conditions. This could explain the less substantial differences between the breeds regarding the fatty acid composition of meat. Maybe other conditions, such as the ages of the animals and different kinds of pasture with or without supplementation, could well affect each breed used here differently. This hypothesis should be considered in future experiments. Anyway, the results of the present investigation established indicators, based on typical productive conditions of Uruguay, about the lipids and fatty acids content of lamb meat for the breeds studied here. Those lipid parameters, which were not determined before, could be used as a baseline for future studies on the nutritional quality of lamb meat produced on pasture in Uruguay.

Conflict of interest

The Authors declare that there is no conflict of interest.

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5.6. Bibliographics References

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6. Discusión general

En esta discusión general abordaremos los aspectos más destacados de los estudios que hicieron parte de esta tesis doctoral, considerando que el objetivo principal fue estudiar las características nutricionales de la carne de corderos de diferentes genotipos, en relación con aquellos nutrientes de gran impacto en la salud humana

6.1. Contenidos minerales

Calcio. En esta investigación llevada adelante en la tesis, de los macrominerales determinados solo el Ca presentó diferencia significativa entre los diferentes genotipos con un rango de a 50 a 67 mg.kg⁻¹ de carne fresca en MD, C y H y un rango de 37 a 44 mg.kg⁻¹ de carne fresca en CPro y MAxC. Según la RDA los requerimientos de Ca de un niño son de 1000 mg/día, lo que significa que el valor más alto de nuestros resultados cubre solo 1.2% de las necesidades diaria. Si bien la carne, no es una fuente importante en calcio para la nutrición humana, este mineral cumple roles claves en la fisiología del músculo (Belhaj et al., 2021; Osorio et al., 2007). El contenido de minerales afecta algunos atributos de calidad de la carne, como el color, la terneza y la oxidación, características organolépticas valoradas por el consumidor (Osorio et al., 2007). En particular, los iones Ca desempeñan un papel fundamental en la determinación de la terneza de la carne, característica de gran importancia para el consumidor (Kemp et al., 2010). El aumento de los niveles de iones de Ca en el músculo y en plasma determina un aumento de la actividad del sistema de las calpaínas, que actúan degradando y autolisando las proteínas musculares y como consecuencia, mejora la terneza de la carne (Kemp et al., 2010). Trabajos realizados por Motter et al. (2021) comparando razas bovinas Brahman y Angus, encontraron diferencias genéticas en la actividad de la calpastatina (antagonista de las calpaínas). Los genotipos de Corriedale Pro y Merino Dohne presentaron diferencias en contenido de Ca ($P<0.05$), podría estar afectando la terneza de la carne (no medido en este trabajo), debido a distinta sensibilidad de los receptores del sistema de calpainas.

calpastatina. Ribeiro et al. (2020) comparó la concentración de calcio en corderos Damara (166,77 mg Ca/kg MS) y Dorper en crecimiento (135,23 mg Ca/kg MS), con 45 kg de peso a la faena, en donde la raza Damara presentó la mayor concentración. La diferencia en la concentración de Ca en el músculo está asociado a la función que cumple en la contracción muscular, y a un efecto de la raza relacionado con las diferentes tasas de crecimiento, ya que los corderos Dorper tienen mayores tasas de crecimiento en comparación con los Damara, priorizando, así, el Ca⁺² para el crecimiento tanto del esqueleto como del músculo (Almeida, 2011; Scanlon et al., 2013). Una asociación entre contenidos de calcio en músculo, terneza de la carne y tasas de crecimiento permitiría asociar los tres genotipos con más Ca (MD, C, H) con los de menor Ca (CPro y MAXC), hay que continuar estudiando.

Zinc. Un interesante aporte de zinc de todos los genotipos estudiados califica a la carne de estos corderos como importante fuente nutricional para la salud humana. El Zn juega un rol importante sobre el sistema inmune de los humanos y su deficiencia puede traer un impacto negativo en el crecimiento de niños y varias enfermedades en adultos (Juarez et al., 2021). Si bien en este trabajo se obtuvieron algunas diferencias entre los genotipos, Corriedale, Corriedale Pro, Highlander® y Merino Dohne destacaron por su mayor aporte a los requerimientos de niños (RDA, 3-5 mg/día) (Food and Nutrition Board, Institute of Medicine [FNB/IOM], 2001), ya que 100 gramos de esta carne, contribuye con el 60-100 % del requerimiento de niños escolares. El menor contenido de zinc se observó en las cruzas (MAXC); si bien es significativamente inferior al de Corriedale, su contribución es cercana al 60 % para una porción de 100 gramos de carne. Según la bibliografía encontrada, el efecto de las líneas genéticas sobre el contenido de zinc en la carne de corderos, estaría asociada principalmente con el tipo de músculo y de fibra muscular presentes en el músculo (Ponnampalam et al., 2019).

Manganoso. La crusa MAXC presentó los menores niveles de Mn (191 mg.kg⁻¹) respecto de los otros cuatro genotipos (C 252, CP 2,82, H 304 y MD 308 mg.kg⁻¹). El rol del Mn en el músculo está muy relacionado con las enzimas antioxidantes; en particular, formando parte de la metaloenzima superóxido dismutasa (Mn-SOD) mitocondrial (Zidenberg-Cherr et al., 1985), lo que podría sugerir que, al

cruzar estos genotipos, podría verse afectada su actividad antioxidante y estar proclive a la peroxidación, aspecto a desarrollar en futuras investigaciones.

Formas de hierro. Corriedale presentó mayor contenido de HFe. Esto estaría determinando que, según Greenwood et al. (2006) y Pannier, Pethick, Boyce et al. (2014) el músculo *longissimus thoracis* tendría mayor número de fibras oxidativas (tipo II rojas) en Corriedale. La raza MD se caracteriza por presentar un crecimiento rápido y carnes más magras (Cottle, 2010), lo que explicaría las diferencias en contenido de Ca, Zn y NHFe. Trabajos previos de Montossi et al. (2014), en donde compararon Corriedale puro con diferentes cruzas CxMD (50 % MD, 75 %MD), muestran que, a medida que aumenta la participación del MD, se obtiene un crecimiento más rápido, mayor peso de corderos a la faena y cortes más magros (mayor tipo de fibras glucolíticas-fibras tipo II) (Greenwood et al., 2006). Merino Dohne se caracteriza por tener una alta tasa de crecimiento (Cottle, 2010), por lo que tendría fibras más blancas por menor cantidad de mioglobina y, en consecuencia, mayor cantidad de NHFe (Knight et al., 2020), como el biotipo MD en nuestros resultados, coincidiendo con las tendencias de Pannier, Pethick, Boyce et al. (2014).

6.2. Grasa intramuscular y ácidos grasos

Grasa intramuscular. La grasa intramuscular (GIM) es beneficiosa en lo que respecta a la calidad de carne porque está asociada a dos características importantes para el consumidor que son el sabor (Nute et al., 2007) y la terneza (Pannier, Pethick, Geesink, et al., 2014). Pero, por otro lado, ha sido cuestionada por estar relacionada, según estudios médicos, con factores predisponentes a enfermedades cardiovasculares, algunos tipos de cáncer y obesidad. La variación de la GIM depende de una compleja interacción entre la genética del animal, la edad, el ambiente donde se produce y sus interacciones (Pannier, Pethick, Geesink, et al., 2014). Lambe et al. (2008) reportaron que seleccionando corderos de la raza Texel por musculatura y reducidos en grasa tuvieron menor cantidad de grasa intramuscular (1,6 %) en el músculo *longissimus* comparando con la raza Scottish Blackface (2,3 %). Estos valores están por debajo de los sugeridos por Hopkins et al. (2016) de 5 % de GIM como valor objetivo para tener

una carne de calidad para el consumo humano, lo que los autores denominan “healthing satisfaction” y de Pannier, Pethick, Geesink, et al. (2014) que reportan valores de GIM máximos para el consumo humano de 3,9 %. Los valores reportados por Lambe et al. (2008), por lo tanto, estarían en valores por debajo de los requeridos valorada positivamente por los consumidores. En nuestros resultados, Highlander® presentó el menor contenido ($2,39 \pm 0,18 \%$) respecto de los otros cinco genotipos estudiados, Merino Dohne, Corriedale, Corriedale Pro, crusa CxMA y Romney Marsh (3,21 %, 3,46 %, 4,17 %, 4,21 % y 4,49 %, respectivamente), sin diferencias entre estas últimas. Esto muestra que H estaría por debajo de los valores de grasa intramuscular que sugieren Pannier, Pethick y Geesink, et al., (2014), no así el resto de los genotipos estudiados.

Ácidos grasos. Con respecto a la composición de ácidos grasos, el impacto de la genética puede estar confundida por las diferencias en el grado de engrasamiento, peso y edad fisiológica a la faena, y el sistema de producción (Fisher et al., 2000; Sañudo et al., 2000). De Smet y Vossen (2016) enfatizan que es la dieta lo que marca la variación del perfil de ácidos grasos de la carne.

Los rumiantes se caracterizan por presentar bajas cantidades de AGPI, debido al proceso de biohidrogenación que se da en el rumen (Wood et al., 2004); pero, por otro lado, permite la incorporación preferencial de ácidos grasos de cadena larga de la familia AGPI n-3 en los glicerofosfolípidos en oposición a los triglicéridos (Bessa et al., 2015). Los productos intermedios de la biohidrogenación permiten la acumulación de ácidos grasos insaturados de cadena larga, ácidos grasos conjugados (ALC) y ácidos grasos monoinsaturados a través de la isomerización y biohidrogenación de los ácidos grasos insaturados de la dieta por enzimas de los microorganismos del rumen (Lee y Jenkins, 2011; Turner et al., 2015). Sumado a esto, los microorganismos del rumen sintetizan de forma endógena ácidos grasos ramificados y saturados de cadena impar que incorporan en sus membranas celulares y luego aparecen en el tejido del animal, lo que sugiere una relación directa con la masa microbiana (Fievez et al., 2012; Vlaeminck et al., 2006). Cuando la dieta es principalmente forraje fresco, como en nuestro trabajo, se genera un ambiente ruminal en donde predominan las bacterias celulolíticas que degradan las fibras, como *Butyrivibrio fibrisolvens*, las cuales en el

proceso de biohidrogenación generan mayor cantidad de compuestos intermediarios como los ácidos vaccénico (VA), ruménico (RA) y ácidos grasos de cadena larga n-3 (LCn-3) (Vlaeminck et al., 2006). Por lo dicho anteriormente, la dieta tiene una fuerte influencia en la masa microbiana y, por tanto, como mencionan los autores anteriores, en los ácidos grasos producidos y presentes en la carne. Pero también, según Zhang et al. (2020), existen diferencias entre individuos y, tal vez, entre razas, en el predominio de determinadas especies de bacterias ruminantes clulolíticas (diferencias en el proceso de biohidrogenación, isomerización), las cuales estarían determinando variación de la composición de ácidos grasos a nivel del tejido muscular. Esto establece un fundamento para la continuación de investigaciones adicionales.

Índices de calidad lipídica (índice de salud). A partir del perfil de los ácidos grasos se calcularon los índices nutricionales y de lípidos saludables de la carne de los diferentes genotipos. La relación AGPI/AGS es importante porque, al aumentarse, reduce el riesgo de enfermedades cardiovasculares. Este índice se utiliza para calcular el factor de riesgo de los alimentos y se aconseja que esta relación sea, como mínimo, de 0,45 para carne (Warris, 2010). Trabajos anteriores encontraron valores de AGPI/AGI en carne de vacuno y cordero entre 0,11 y 0,15 (Geay et al., 2001) y de 0,10 a 0,15 en corderos de diferentes cruzas raciales (Hoffman et al., 2003). En nuestro experimento, la relación AGPI/AGS varió entre 0,10 y 0,14, encontrándose por debajo del valor recomendado, pero dentro de los valores reportados por la bibliografía. Otro índice está relacionado con el anterior es la relación n6:n3, ya que el aumento del contenido de ácido alfa linolénico (C18:3 n-3) un AGPI, característico en productos de origen animal con dieta forrajera, reducirá esta última relación. Por ello, se recomiendan valores inferiores a 4 (De Lemos et al., 2017) o incluso entre 1:1 y 2:1 (Simopoulos, 2010) para esta relación, ya que se asocia con la aparición de arteriosclerosis y problemas cardiovasculares (Simopoulos, 2002; Wood et al., 2003). Toda la carne de los animales evaluados en nuestro trabajo presenta una proporción n-6:n-3, acorde a la recomendación de 2:1 (Simopoulos, 2010). Sin embargo, hoy en día, esa relación se cuestiona respecto a su real utilidad para la salud humana (Harris, 2018). Los índices utilizados en nuestro trabajo para evaluar la protección potencial frente a las enfermedades cardiovasculares fueron el IA, el IT y el h/H (Chen y Liu,

2020). El valor de IA (cuyo valor recomendado es el más bajo posible) observado en nuestro trabajo osciló entre 0,63 y 0,89. Estos valores fueron ligeramente superiores a los reportados por Cadavez et al. (2020), Belhadj et al. (2020) y el trabajo de compilación del Programa Cooperativo para el Desarrollo Tecnológico Agroalimentario y Agroindustrial del Cono Sur-Instituto Interamericano de Cooperación para la Agricultura [PROCISUR-IICA] (2015). Los valores de IT observados en nuestro trabajo oscilaron entre 1,51 y 1,80. Estos valores son superiores a los reportados por Cadavez et al. (2020), Belhaj et al. (2020) y la compilación de PROCISUR-IICA (2015), en donde el valor recomendado debe ser lo más bajo posible, para reducir los efectos trombogénicos en humanos (Bozbas et al., 2021). En estos dos índices no hubo diferencias estadísticas entre los diferentes genotipos, lo que podría deberse al peso vivo a la faena. Los valores los índices h/H observados en nuestro trabajo oscilaron entre 1,64 y 2,12 y el C mostró un índice superior al MD. Para este índice, el valor recomendado en la carne debe ser lo más alto posible para minimizar el riesgo de hipercolesterolemia, que conduce a enfermedades cardiovasculares (Moussavi Javardi et al., 2020). Algunos de los índices presentados, como P/S y TI, no estuvieron dentro de los valores recomendados, por lo que debe investigarse más para intentar mejorar estos parámetros.

Se han calculado también, índices enzimáticos para las enzimas desaturasas delta 9, delta 5 y delta 6, la elongasa y la tioesterasa en un intento de detectar diferencias en el metabolismo de los lípidos entre los genotipos estudiados en el presente trabajo. Este procedimiento se ha utilizado también en el campo médico como forma sencilla de evaluar las actividades de enzimas como las desaturasas, elongasas y tioesterasas en algunas patologías humanas (Czumaj y Sledzinski, 2020; Do et al., 2011; Ebbesson et al., 2012). En nuestro trabajo se pudo observar que C, CPRO y CxAM mostraron una actividad de la delta 9-C16 mayor que MD. Este resultado podría explicar las diferencias en el contenido de C16:1, indicado para los glicerolípidos, para CPRO y CxAM, pero no para C. En el caso de la enzima delta 9-C18, así como en la suma de las actividades de ambas enzimas delta 9-C16+C18, no existieron diferencias ($P > 0,05$) entre los biotipos estudiados. La enzima delta 9 introduce un doble enlace *cis* en la posición D9 entre los carbonos 9 y 10, y los sustratos preferidos son palmitoil-CoA

para delta 9-C16 y estearoil-CoA para delta 9-C18, que conducen a su conversión en palmitoleoil-CoA y oleoil-CoA, respectivamente (Ntambi y Miyazaki, 2003). Los AGMI y, en particular, los C18:1 son de gran importancia para la estructura y función de las membranas basadas en fosfoglicéridos (Liu et al., 2011; Santa-María et al., 2023). Sin embargo, globalmente no se observó una diferencia clara entre los genotipos en cuanto a la síntesis *de novo* y la deposición de AGMI, tanto en glicerolípidos como en glicerofofolípidos, en el músculo *longissimus thoracis*. El hecho de que los animales hayan sido alimentados con la misma dieta forrajera y el mismo manejo podría haber minimizado las posibles diferencias de expresión de enzimas entre los genotipos. Los índices enzimáticos para delta 5 tampoco mostraron diferencias entre los animales, mientras que, para delta 6, los C presentaron una mayor actividad que los H. Este resultado estaría explicando el mayor contenido en C18:3n3 para C en comparación con H, en los glicerolípidos en contraste con los glicerofosfolípidos. En el caso de las actividades de la elongasa, C mostró una mayor actividad en comparación con MD. Las enzimas elongasas añaden dos átomos de carbono al ácido graso C16:0 y se forma el ácido graso C18:0, pero también elongan otros ácidos grasos a partir de los dos ácidos grasos esenciales C18:2n6 y C18:3n3 (Lara et al., 2018; Raes et al., 2004). Sin embargo, la comparación entre C y MD en cuanto al contenido de C18:0 y AGPI no mostraron diferencias en los glicerolípidos ni en los glicerofosfolípidos. Es posible que se deba a que, la actividad diferencial observada de las elongasas entre C y MD fuera muy pequeña como para afectar de forma significativa al contenido de C18:0 y AGPI en la carne de ambos genotipos. Al igual que en el caso de las desaturasas, el hecho de que los animales hayan sido criados en las mismas condiciones podría haber minimizado las diferencias entre genotipos en cuanto a las actividades de las elongasas. Con respecto al índice de tioesterasa, no hubo diferencias entre los seis biotipos estudiadas. La tioesterasa forma parte del complejo enzimático ácido graso sintasa codificado por el gen de la sintasa de ácidos grasos (FASN) en mamíferos y regula principalmente la formación de C16:0 como producto final y C14:0 como producto menor (Pewan et al., 2020; Popova et al., 2020). Teniendo en cuenta, como se ha informado anteriormente, la implicación del C16:0 en las enfermedades cardiovasculares en humanos, podría ser interesante investigar cómo

se puede modular la actividad tioesterasa en la carne de cordero, lo que podría ayudar a mejorar la calidad nutricional y sanitaria de la carne.

7. Conclusión General

La carne de corderos de los genotipos estudiados producidos en sistema pastoril presenta una riqueza en nutrientes de alto valor, como los elementos traza, hierro hemo, vitamina B12 y ácidos grasos, que la caracterizan como un alimento valioso y adecuado a los requerimientos nutricionales de la mayoría de las personas. Desde una perspectiva de los genotipos, surgen asociaciones significativas con los minerales Mn, Ca y Zn, así como con las cantidades de HFe y NHFe, que permitirían valorizar la carne de determinado genotipo en función de objetivos comerciales o demandas específicas, o asociar la presencia de mayor contenido de nutrientes con la composición muscular y de fibras de cada genotipo.

Con respecto al perfil de ácidos grasos, los resultados del estudio muestran una superposición entre los biotipos genéticos, relacionados con el metabolismo lipídico composicional, excepto para algunos ácidos grasos de importancia como C16:0, C18:3n3 y CLA para glicerolípidos y C18:1, C18:2n6 y C18:3n3 para glicerofosfolípidos. Asimismo, se encontraron otras diferencias en relación a BCFAai, h/H y de la actividad enzimática de Δ-9-C16, Δ-6 y elongasa, que no se dio en todos los biotipos genéticos estudiados, y puede ser una base para nuevos estudios. Igualmente, es de destacar que los biotipos genéticos estudiados presentan buenos indicadores nutricionales lipídicos en comparación con los resultados de otras investigaciones en corderos, excepto para los ácidos grasos EPA y DHA, ya que estos biotipos presentan un contenido bajo cuando la dieta es únicamente pastoril a base de gramíneas y leguminosas, en comparación con la literatura científica, en la cual se utilizan suplementos ricos en lípidos poliinsaturados.

Los resultados de esta tesis justifican y dan margen para seguir investigando sobre la calidad de carne de corderos en lo que respecta a la composición mineral y de ácidos grasos y las posibles interacciones con diferentes dietas atendiendo a lograr mayores contenidos de diferentes nutrientes para obtener productos cárnicos valorizados y enriquecidos.

8. Bibliografía general

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