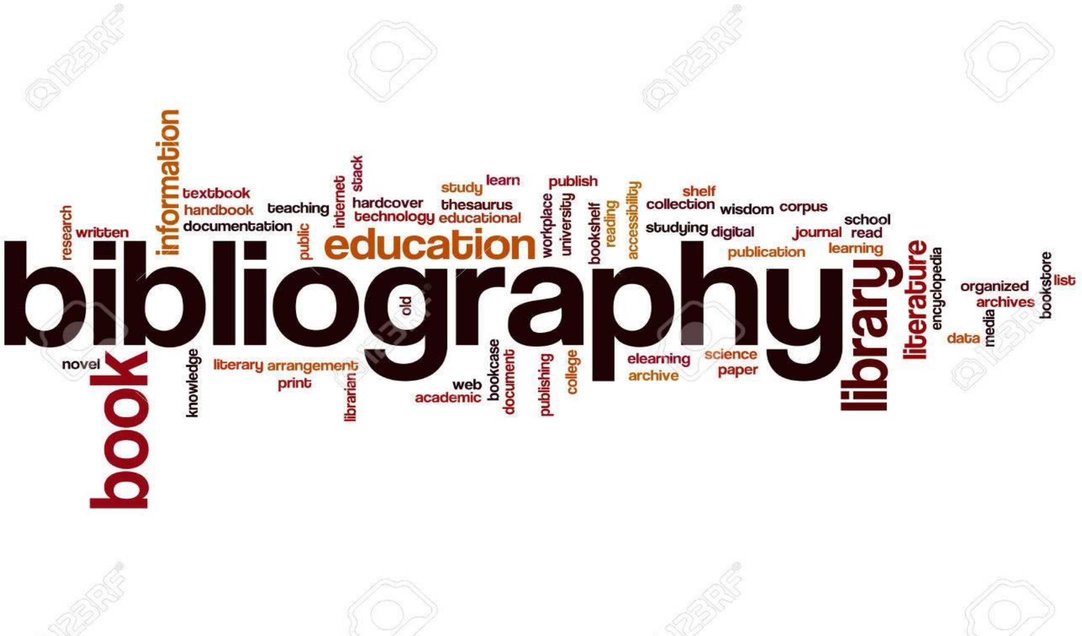


Bibliografía Científica

Anexo 2021

SERIDA
Servicio Regional de Investigación
y Desarrollo Agroalimentario

Unidad de Archivo y Biblioteca

[Ana Rodríguez Navarro, marzo 2022]

PRESENTACIÓN

Uno de los resultados más visibles de la actividad científica de una institución son las publicaciones de sus miembros y colaboradores en revistas científicas de prestigio internacional. En este contexto, la Bibliografía que ahora se presenta, recopila la producción científica del SERIDA en el año 2021 que aparece recogida en las diversas bases de datos multidisciplinares y de carácter internacional que integran la *Web of Science* (WoS).

La *Web of Science* permite, por una parte, la recuperación de los trabajos del SERIDA indexados en las revistas científicas de mayor visibilidad e impacto y por otra, su caracterización desde el punto de vista bibliométrico. El objetivo ha sido recuperar todos los artículos con al menos un autor perteneciente al SERIDA, por lo que se definió una estrategia de búsqueda usando el campo de "afiliación", una vez que fue ratificada por la entidad gestora de la base de datos, la propuesta de consolidación de las distintas variantes del nombre de la entidad. El resto de opciones fueron las establecidas por defecto en la interfaz de búsqueda, excepto el período que se acotó al año 2021, aceptándose además para su inclusión en la bibliografía, aquellos registros que tuvieran fecha de "acceso anticipado" en 2021, aunque la fecha de publicación definitiva sea de un año posterior. Cuando se produce este hecho, ambas fechas, tanto la de acceso anticipado, como la de publicación definitiva, han quedado reflejadas en las respectivas entradas bibliográficas y lógicamente no volverán a ser incluidas en la bibliografía del próximo año.

Esta nueva actualización se compone de 66 artículos y la recopilación se ha organizado y redactado en el estilo bibliográfico APA (7ª ed.), que asocia autor y año por orden alfabético. Cada referencia, se ha completado con el resumen o abstract del artículo, la indicación de área o áreas del SERIDA a la que pertenecen los investigadores, y el enlace al artículo.

El repertorio se ha enriquecido con varios índices y gráficos: un índice alfabético de los autores que pertenecen al SERIDA (Índice A), un índice alfabético de los 45 títulos de las revistas en las que aparecen publicados los artículos, con indicación del factor de impacto de cada una de ellas y del cuartil de importancia al que pertenecen según el *Journal Citation Reports* (JCR) (Índice B), y un índice de los artículos según el área de investigación del SERIDA al que se adscriben los investigadores (Índice C).

Las figuras, por su parte, pretenden resaltar gráficamente los porcentajes de producción según las áreas de investigación del SERIDA (Figura 1), los países y las instituciones con las que se ha colaborado (Figuras 2 y 3 respectivamente, los títulos de las revistas en las que más se ha publicado (Figura 4), y la importancia de la revista en la que aparecen publicados los artículos (Figuras 5 y 6).

Sistematizando y difundiendo los resultados de la investigación del centro, creemos que la biblioteca contribuye de forma eficaz a aumentar la visibilidad de la institución y de sus investigadores puesto que además el repertorio se publicará, como en ocasiones anteriores en el *Repositorio Institucional de Asturias* (RIA).

LISTADO DE ABREVIATURAS

^{a}	Fecha de acceso anticipado
CHF	Área de Cultivos Hortofrutícolas y Forestales
EDA	Área de Experimentación y Demostración Agroforestal
GRA	Área de Genética y Reproducción Animal
NPF	Área de Nutrición, Pastos y Forrajes
SA	Área de Sanidad Animal
SRA	Área de Selección y Reproducción Animal
SPA	Área de Sistemas de Producción Animal
TA	Área de Tecnología de los Alimentos
OA	Acceso Abierto
^{p}	Fecha de publicación

LISTADO DE REFERENCIAS

1.- ADALID, R., FELIU, C., SOMOANO, A., MIÑARRO, M., VENTURA, J., TORRES, J., MIQUEL, J., & FUENTES, M.V. (2021). Ecological analysis of the helminth community of *Microtus lusitanicus* (Gerbe, 1879) (Rodentia) in Asturias (NW Spain). *Animals 11*, Art. 3055. DOI: 10.3390/ani11113055. (OA) [CHF] [Ver online]

Abstract: The Lusitanian pine vole, *Microtus lusitanicus*, an endemic fossorial rodent of the Iberian Peninsula, has a burrowing behaviour and prefers to live underground. It feeds on bark and roots causing severe damage to trees. In Asturias (NW Spain), where *M. lusitanicus* is considered a pest in several orchards, a faunistic-ecological study was carried out to describe the helminth community of this species and the main factors that could influence its helminth component species. For this purpose, our own collection of 710 voles from several orchards of various locations in Asturias was used. Eight helminth species, four cestodes and four nematodes, were found. Statistical non-parametric tests were used to analyse the effects of extrinsic and intrinsic factors on the diversity of the helminth community and species prevalence and abundance. The results show the influence of climate variables, the year and season of capture, as well as host age, on the diversity of the helminth community and the infection parameters of some helminth species, underlining the importance of their life cycles. In addition to shedding light on the helminth community of this rodent in Asturias, the results obtained could be used to improve the biological methods applied to fight the *M. lusitanicus* pest.

2.- ALLEN-PERKINS A., …, M. MIÑARRO…, BARTOMEUS, I. (180 autores) (2021^{a}- 2022^{p}). CropPol: a dynamic, open and global database on crop pollination. *Ecology,103*(3), Art. 3614. DOI:10.1002/ecy, 3614. (OA) [CHF] [Ver online]

Abstract: Seventy five percent of the world's food crops benefit from insect pollination. Hence, there has been increased interest in how global change drivers impact this critical ecosystem service. Because standardized data on crop pollination are rarely available, we are limited in our capacity to understand the variation in pollination benefits to crop yield, as well as to anticipate changes in this service, develop predictions, and inform management actions. Here, we present CropPol, a dynamic, open, and global database on crop pollination. It contains measurements recorded from 202 crop studies, covering 3,394 field observations, 2,552 yield measurements (i.e., berry mass, number of fruits, and fruit density [kg/ha], among others), and 47,752 insect records from 48 commercial crops distributed around the globe. CropPol comprises 32 of the 87 leading global crops and commodities that are pollinator dependent. *Malus domestica* is the most represented crop (32 studies), followed by *Brassica napus* (22 studies), *Vaccinium corymbosum* (13 studies), and *Citrullus lanatus* (12 studies). The most abundant pollinator guilds recorded are honey bees (34.22% counts), bumblebees (19.19%), flies other than Syrphidae and Bombyliidae (13.18%), other wild bees (13.13%), beetles (10.97%), Syrphidae (4.87%), and Bombyliidae (0.05%). Locations comprise 34 countries distributed among Europe (76 studies), North America (60), Latin America and the Caribbean (29), Asia (20), Oceania (10), and Africa (7). Sampling spans three decades and is concentrated on 2001–2005 (21 studies), 2006–2010 (40), 2011–2015 (88), and 2016–2020 (50). This is the most comprehensive open global data set on measurements of crop flower visitors, crop pollinators and pollination to date, and we encourage researchers to add more datasets to this database in the future. This data set is released for non-commercial use only. Credits should be given to this paper (i.e., proper citation), and the products generated with this database should be shared under the same license terms (CC BY-NC-SA).

3.- ÁLVAREZ, I., FERNÁNDEZ, I., TRAORE, A., MENÉNDEZ-ARIAS, N. A., & GOYACHE, F. (2021). Population structure assessed using microsatellite and SNP data: An empirical comparison in West African cattle. *Animals, 11*(1), Art. 151. DOI: 10.3390/ani11010151. (OA) [GRA] [Ver online]

Abstract: A sample of 185 West African cattle belonging to nine different taurine, sanga, and zebu populations was typed using a set of 33 microsatellites and the BovineHD BeadChip of Illumina. The information provided by each type of marker was summarized via clustering methods and principal component analyses (PCA). The aim was to assess differences in performance between both marker types for the identification of population structure and the projection of genetic variability on geographical maps. In general, both microsatellites and Single Nucleotide Polymorphism (SNP) allowed us to differentiate taurine cattle from zebu and sanga cattle, which, in turn, would form a single population. Pearson and Spearman correlation coefficients computed among the admixture coefficients (fitting K = 2) and the eigenvectors corresponding to the first two factors identified using PCA on both microsatellite and SNP data were statistically significant (most of them having $p < 0.0001$) and high. However, SNP data allowed for a better fine-scale identification of population structure within taurine cattle: Lagunaire cattle from Benin were separated from two different N'Dama cattle samples. Furthermore, when clustering analyses assumed the existence of two parental populations only (K = 2), the SNPs could differentiate a different genetic background in Lagunaire and N'Dama cattle. Although the two N'Dama cattle populations had very different breeding histories, the microsatellite set could not separate the two N'Dama cattle populations. Classic bidimensional dispersion plots constructed using factors identified via PCA gave different shapes for microsatellites and SNPs: plots constructed using microsatellite polymorphism would suggest the existence of weakly differentiated, highly intermingled, subpopulations. However, the projection of the factors identified on synthetic maps gave comparable images. This would suggest that results on population structuring must be interpreted with caution. The geographic projection of genetic variation on synthetic maps avoids interpretations that go beyond the results obtained, particularly when previous information on the analyzed populations is scant. Factors influencing the performance of the projection of genetic parameters on geographic maps, together with restrictions that may affect the election of a given type of markers, are discussed.

4.- ÁLVAREZ, S., MÉNDEZ, P., & MARTÍNEZ-FERNÁNDEZ, A. (2021). Silage fermentation and chemical composition of *Chamaecitysus proliferus* var *palmensis* (Tagasaste) and *Pennisetum* sp (Maralfalfa) using different additives. *Journal of Animal and Plant Sciences-Japs*, *31*(4), 929-936. DOI: 10.36899/japs.2021.4.0286. (OA) [NPF] [Ver online]

Abstract: The aim of this study was to evaluate the fermentation characteristics and nutritive value of two interesting forage species (maralfalfa and tagasaste) ensiled with molasses and beet pulp as additives. Ensilability parameters and chemical composition were analysed in two different stages of growth in maralfalfa (flowering and vegetative growing) and two different branch diameters in tagasaste (branches > 5mm, branches ≤ 5mm). According to ensilability results, four combinations with additives (two for tagasaste and two for maralfalfa) were ensiled at laboratory scale microsilos, in order to evaluate fermentative and nutritive parameters in two different conservation periods (35d and 70d). Tagasaste showed high dry matter (DM) content and fermentability ratio (FR) but a very low sugar content (WSC). Maralfalfa showed poor initial ensiling characteristics, low DM and WSC and high BC (buffering capacity). Supplementing tagasaste with molasses improves the fermentative composition in mini-silos with adequate ammonia-N and lactic acid concentrations detected. Moreover, the addition of

beet pulp and molasses to the ensiled maralfalfa allows achieving the pH value under 4 and NH3-N content below 10%, with an interesting content of lactic acid improving the fermentation quality of silage.

5.- BAIZÁN, S., VICENTE, F., & MARTÍNEZ-FERNÁNDEZ, A. (2021). Management influence on the quality of an agricultural soil destined for forage production and evaluated by physico-chemical and biological indicators. *Sustainability, 13*(9), Art. 5159. DOI: 10.3390/su13095159. (OA) [NPF] [Ver online]

Abstract: The European Common Agricultural Policy promotes the sustainable use of soils through the principle of cross-compliance that links direct payments to good farming practices. Thus, it is necessary to find sustainable alternatives to the conventional management for forage production in the Atlantic Arc dairy farms. Two alternative managements (faba bean in monoculture—FB—and faba bean–Italian ryegrass intercrop—FBIR) were cultivated with organic fertilization during two consecutive years, and compared to the conventional management (Italian ryegrass in monoculture—IR—under chemical fertilization) as winter crops. Maize was used as the summer crop to complete the rotations using organic and chemical fertilization, respectively. The forage yield of winter fodder was quantified. Soil samples and summer harvests were performed to analyse the physico-chemical and biological parameters. The best forage yield corresponded to FBIR with 9.2 t dry matter (DM) ha^{-1} vs. 7.2 and 5.7 t DM ha^{-1} for FB and IR, respectively. The soil organic matter did not show significant differences among treatments, suggesting that it may be a poor indicator of the management influence on edaphic quality in the short term. Biological indicators were more sensitive and faster to differentiate among managements than chemical indicators. Earthworm abundance was higher in FB and FBIR than IR ($p < 0.001$), and consequently, soil infiltration was improved with the alternative management (13.90 vs. 2.08 and 0.90 min for IR, FB and FBIR, respectively, $p < 0.01$). As a result, the soil health diagnosis of the agroecosystem was better for alternative management.

6.- BELLUCCI, E., MARIO AGUILAR, O., ALSEEKH, S., BETT, K., BREZEANU, C., COOK, D., DE LA ROSA, L., DELLEDONNE, M., DOSTATNY, D.F., FERREIRA, J.J., GEFFROY, V., GHITARRINI, S., KROC, M., KUMAR AGRAWAL, S., LOGOZZO, G., MARINO, M., MARY-HUARD, T., MCCLEAN, P., MEGLIČ, V., MESSER, T., MUEL, F., NANNI, L., NEUMANN, K., SERVALLI, F., STRĂJERU, S., VARSHNEY, R.K., VASCONCELOS, M.W., ZACCARDELLI, M., ZAVARZIN, A., BITOCCHI, E., FRONTONI, E., FERNIE, A.R., GIOIA, T., GRANER, A., GUASCH, L., PROCHNOW, L., OPPERMANN, M., SUSEK, K., TENAILLON, M. & PAPA, R. . (2021). The INCREASE project: Intelligent collections of food-legume genetic resources for European agrofood systems. *Plant Journal, 148*(3), 646-660. DOI: 10.1111/tpj.15472.(OA) [CHF] [Ver online]

Abstract: Food legumes are crucial for all agriculture-related societal challenges, including climate change mitigation, agrobiodiversity conservation, sustainable agriculture, food security and human health. The transition to plant-based diets, largely based on food legumes, could present major opportunities for adaptation and mitigation, generating significant co-benefits for human health. The characterization, maintenance and exploitation of food-legume genetic resources, to date largely unexploited, form the core development of both sustainable agriculture and a healthy food system. INCREASE will implement, on chickpea (*Cicer arietinum*), common bean (*Phaseolus vulgaris*), lentil (*Lens culinaris*) and lupin (*Lupinus albus* and *L. mutabilis*), a new approach to conserve, manage and characterize genetic resources. *Intelligent*

Collections, consisting of nested core collections composed of single-seed descent-purified accessions (i.e., inbred lines), will be developed, exploiting germplasm available both from genebanks and on-farm and subjected to different levels of genotypic and phenotypic characterization. Phenotyping and gene discovery activities will meet, via a participatory approach, the needs of various actors, including breeders, scientists, farmers and agri-food and non-food industries, exploiting also the power of massive metabolomics and transcriptomics and of artificial intelligence and smart tools. Moreover, INCREASE will test, with a citizen science experiment, an innovative system of conservation and use of genetic resources based on a decentralized approach for data management and dynamic conservation. By promoting the use of food legumes, improving their quality, adaptation and yield and boosting the competitiveness of the agriculture and food sector, the INCREASE strategy will have a major impact on economy and society and represents a case study of integrative and participatory approaches towards conservation and exploitation of crop genetic resources.

7.- BLANCO VÁZQUEZ, C., BALSEIRO, A., ALONSO-HEARN, M., JUSTE, R. A., IGLESIAS, N., CANIVE, M., & CASAIS, R. (2021). Bovine Intelectin 2 expression as a biomarker of paratuberculosis disease progression. *Animals*, *11*(5), Art. 1370. DOI: 10.3390/ani11051370. (OA) [SA] [Ver online]

Abstract: Paratuberculosis (PTB), a chronic granulomatous enteritis caused by *Mycobacterium avium* subsp. *paratuberculosis* (MAP), is responsible for important economic losses in the dairy industry. Our previous RNA-sequencing (RNA-Seq) analysis showed that bovine intelectin 2 (ITLN2) precursor gene was overexpressed in ileocecal valve (ICV) samples of animals with focal (log2 fold-change = 10.6) and diffuse (log2 fold-change = 6.8) PTB-associated lesions compared to animals without lesions. This study analyzes the potential use of ITLN2, a protein that has been described as fundamental in the innate immune response to infections, as a biomarker of MAP infection. The presence of ITLN2 was investigated by quantitative immunohistochemical analysis of ICV samples of 20 Holstein Friesian cows showing focal (n = 5), multifocal (n = 5), diffuse (n = 5) and no histological lesions (n = 5). Significant differences were observed in the mean number of ITLN2 immunostained goblet and Paneth cells between the three histopathological types and the control. The number of immunolabelled cells was higher in the focal histopathological type (116.9 ± 113.9) followed by the multifocal (108.7 ± 140.5), diffuse (76.5 ± 97.8) and control types (41.0 ± 81.3). These results validate ITLN2 as a post-mortem biomarker of disease progression.

8.- BLANCO VÁZQUEZ, C., BARRAL, T. D., ROMERO, B., QUEIPO, M., MEREDIZ, I., QUIRÓS, P., ARMENTEROS, J. A., JUSTE, R., DOMÏNGUEZ, L., DOMÏNGUEZ, M., CASAIS, R., & BALSEIRO, A. (2021). Spatial and temporal distribution of *Mycobacterium tuberculosis* complex infection in Eurasian Badger (*Meles meles*) and cattle in Asturias, Spain. *Animals, 11*(5), Art. 1294. DOI: 10.3390/ani11051294. (OA) [SA] [Ver online]

Abstract: The present work investigated the prevalence, spatial distribution, and temporal distribution of tuberculosis (TB) in free-ranging Eurasian badgers (*Meles meles*) and cattle in Asturias (Atlantic Spain) during a 13-year follow-up. The study objective was to assess the role of badgers as a TB reservoir for cattle and other sympatric wild species in the region. Between 2008 and 2020, 673 badgers (98 trapped and 575 killed in road traffic accidents) in Asturias were necropsied, and their tissue samples were cultured for the *Mycobacterium tuberculosis* complex (MTC) isolation. Serum samples were tested in an in-house indirect P22 ELISA to detect antibodies against the MTC. In parallel, data on MTC isolation and single intradermal tuberculin test results were extracted for cattle that were tested and culled as part of the Spanish National Program for the Eradication of Bovine TB. A total of 27/639 badgers (4.23%) were positive for MTC based on bacterial isolation, while 160/673 badgers (23.77%) were found to be positive with the P22 ELISA. The rate of seropositivity was higher among adult badgers than subadults. Badger TB status was spatially and temporally associated with cattle TB status. Our results cannot determine the direction of possible interspecies transmission, but they are consistent with the idea that the two hosts may exert infection pressure on each other. This study highlights the importance of the wildlife monitoring of infection and disease during epidemiological interventions in order to optimize outcomes

9.- CAAMAÑO, J. N., TAMARGO, C., PARRILLA, I., MARTÍNEZ-PASTOR, F., PADILLA, L., SALMAN, A., FUEYO, C., FERNÁNDEZ, A., MERINO, M.J., IGLESIAS, T., & HIDALGO, C.O. (2021). Post-thaw sperm quality and functionality in the autochthonous pig breed Gochu Asturcelta. *Animals, 11*(7), Art. 1885. DOI: 10.3390/ani11071885. (OA) [SRA] [Ver online]

Abstract: Genetic resource banks (GRB) preserve the genetic material of endangered, valuable individuals or genetically relevant breeds. Semen cryopreservation is a crucial technique to reach these goals. Thus, we aimed to assess the sperm parameters of semen doses from the native pig breed Gochu Asturcelta stored at the GRB of Principado de Asturias (GRB-PA, Gijón, Spain), focusing on intrinsic and extrinsic (boar, season) factors. Two straws per boar (n = 18, 8–71 months of age) were thawed, pooled, and assessed after 30 and 150 min at 37 °C by CASA (computer-assisted sperm analysis system; motility and kinematic parameters) and flow cytometry (viability, acrosomal status, mitochondrial activity, apoptosis, reactive oxygen species, and chromatin status). The effects of age, incubation, and season on post-thawing quality were determined using linear mixed-effects models. Parameters were on the range for commercial boar breeds, with chromatin status (SCSA: fragmentation and immaturity) being excellent. Incubation decreased sperm quality and functionality. The boar age did not have a significant effect ($p > 0.05$), but the between-boar variability was significant ($p < 0.001$). The season significantly affected many parameters (motility, kinematics, viability, acrosomal status, mitochondrial activity), especially after 150 min of incubation. In general, samples collected in spring and summer showed higher quality post-thawing, the lowest in winter. In conclusion, the sperm doses from the Gochu Asturcelta breed stored at the GRB-PA showed excellent chromatin status and acceptable characteristics after thawing. Therefore, boar and seasonal variability in this autochthonous breed could be relevant for cryobank management.

10.- CALVETE-TORRE, I., MUÑOZ-ALMAGRO, N., PACHECO, M. T., ANTÓN, M. J., DAPENA, E., RUÍZ, L., MARGOLLES, A., VILLAMIEL, M., & MORENO, F. J. (2021). Apple pomaces derived from mono-varietal Asturian ciders production are potential source of pectins with appealing functional properties. *Carbohydrate Polymers, 264*, Art. 117980. DOI: 10.1016/j.carbpol.2021.117980. (OA) [CHF] [Ver online]

Abstract: Comprehensive chemical characterization of nine mono-varietal apple pomaces obtained from the production of ciders with PDO is described. They were rich in essential minerals, fibers (35–52.9 %), and polyphenols. High levels in GalA (11.8–21.6 %), revealed the suitability of these apple pomaces as efficient sources of pectins. Extracted pectins showed high variability in monomer composition, with degrees of methylesterification, strongly associated with pectins functional properties, ranging from 58 to 88 %. For a subset of apple pomace varieties, pectin extraction was accomplished by conventional acid heat treatment or ultrasound. Despite ultrasound-assisted extraction did not improve pectin yield, it minimized levels of “non-pectin” components as revealed by the low content of Glc/Man, leading to the obtainment of high-purity pectin. Our work highlights the key role played by the selection of the apple variety to streamline the potential food applications (gelling/thickening agents or prebiotics) of the extracted pectins that largely depend on their structural features.

11.- CAMPA, A., RODRÍGUEZ MADRERA, R., SUÁREZ VALLES, B., & FERREIRA, J. J. (2021). Variation of morphological, agronomic and chemical composition traits of local hazelnuts collected in northern Spain. *Frontiers in Plant Science, 12,* Art. 659510. DOI: 10.3389/fpls.2021.659510. (OA) [CHF] [TA] [Ver online]

Abstract: Hazelnut is a traditional crop in northern Spain, where it grows wild as well as being cultivated. A field collection of 41 local and 17 non-local accessions, including 15 well-known cultivars, was established at SERIDA in Villaviciosa, Spain. Here, phenotypic variation was documented for phenological and morphological traits and chemical composition. A large degree of variation for most morphological and phenological traits, except nut maturity date, was revealed. Estimates of broad-sense heritability were high (>0.75) for most of the assessed characters, except for the first male bloom date (0.65), male and female flowering periods (0.40, 0.31), kernel weight (0.69), and kernel percentage (0.33). Local accessions produced smaller nuts and kernels than well-known cultivars but with higher kernel percentage. Limited overlapping between the male and female flowering periods (dychogamy) was observed, except for 'Forcinas 1', 'Forcinas 2', and 'Morell'. The local accessions generally exhibited significantly later male and female flowering compared with the reference cultivars. The local materials showed similar nutritional values to those reported previously for hazelnut. Moreover, the local accessions presented average values similar to the non-local accessions for total fat, ash and carbohydrate contents, as well as energy value, but their protein contents were lower. Their oils were rich in functional compounds, such as unsaturated fatty acids (average: 90.1%), tocopherols (514 mg/kg) and squalene (294.3 mg/kg). A hierarchical clustering on principal components analysis grouped the accessions and differentiated eight local accessions from the rest, including the landrace 'Casina'. This finding provides potential new cultivars, as well as sources of desirable traits, for European hazelnut breeding programs.

12.- CANIVE, M., FERNÁNDEZ-JIMENEZ, N., CASAIS, R., VÁZQUEZ, P., LAVIN, J. L., BILBAO, J. R., BLANCO-VÁZQUEZ, C., GARRIDO, J. M., JUSTE, R. A., & ALONSO-HEARN, M. (2021). Identification of loci associated with susceptibility to bovine paratuberculosis and with the dysregulation of the MECOM, eEF1A2, and U1 spliceosomal RNA expression. *Scientific Reports, 11*(1), Art. 313. DOI: 10.1038/s41598-020-79619-x. (OA) [SA] [Ver online]

Abstract: Although genome-wide association studies have identified single nucleotide polymorphisms (SNPs) associated with the susceptibility to *Mycobacterium avium* subsp. *paratuberculosis* (*MAP*) infection, only a few functional mutations for bovine paratuberculosis (PTB) have been characterized. Expression quantitative trait loci (eQTLs) are genetic variants typically located in gene regulatory regions that alter gene expression in an allele-specific manner. eQTLs can be considered as functional links between genomic variants, gene expression, and ultimately phenotype. In the current study, peripheral blood (PB) and ileocecal valve (ICV) gene expression was quantified by RNA-Seq from fourteen Holstein cattle with no lesions and with PTB-associated histopathological lesions in gut tissues. Genotypes were generated from the Illumina LD EuroG10K BeadChip. The associations between gene expression levels (normalized read counts) and genetic variants were analyzed by a linear regression analysis using *R Matrix eQTL 2.2.* This approach allowed the identification of 192 and 48 *cis*-eQTLs associated with the expression of 145 and 43 genes in the

PB and ICV samples, respectively. To investigate potential relationships between these *cis*-eQTLs and MAP infection, a case–control study was performed using the genotypes for all the identified *cis*-eQTLs and phenotypical data (histopathology, ELISA for MAP-antibodies detection, tissue PCR, and bacteriological culture) of 986 culled cows. Our results suggested that the heterozygous genotype in the *cis*-eQTL-rs43744169 (T/C) was associated with the up-regulation of the MDS1 and EVI1 complex (*MECOM*) expression, with positive ELISA, PCR, and bacteriological culture results, and with increased risk of progression to clinical PTB. As supporting evidence, the presence of the minor allele was associated with higher *MECOM* levels in plasma samples from infected cows and with increased MAP survival in an ex-vivo macrophage killing assay. Moreover, the presence of the two minor alleles in the *cis*-eQTL-rs110345285 (C/C) was associated with the dysregulation of the eukaryotic elongation factor 1-α2 (*eEF1A2*) expression and with increased ELISA (OD) values. Finally, the presence of the minor allele in the *cis*-eQTL rs109859270 (C/T) was associated with the up-regulation of the U1 spliceosomal RNA expression and with an increased risk of progression to clinical PTB. The introduction of these novel functional variants into marker-assisted breeding programs is expected to have a relevant effect on PTB control.

13.- CIORDIA, M., LOUREIRO, M. D., & GONZALEZ, A. J. (2021). First report of *Neofusicoccum parvum* causing canker on *Castanea sativa* in Spain. *Plant disease*. DOI: 10.1094/pdis-06-21-1231-pdn. (OA) [CHF] [TA] [Ver online]

Abstract: In April 2021, depressed bark with dark-reddish coloration was observed on the stem of a 5-year-old chestnut (*Castanea sativa* Mill.) plant, acquired from a commercial Galician nursery. One tissue sample was collected from the injury of this plant, surface sterilized with 96% ethanol for 30 s, dried on sterilized tissue paper, plated on potato dextrose agar (PDA), and incubated at 25°C. Fungal colonies were consistently isolated and, after 5 days, developed abundant greyish-white aerial mycelium. Two weeks later, pycnidia with fusiform conidia were observed.

14.- COTO-MONTES, A., GONZALEZ-BLANCO, L., ANTUÑA, E., MENENDEZ-VALLE, I., BERMEJO-MILLO, J. C., CABALLERO, B., VEGA NAREDO, I., POTES, Y. (2021). The interactome in the evolution from frailty to sarcopenic dependence. *Frontiers in Cell and Developmental Biology, 9*, Art. 792825. DOI: 10.3389/fcell.2021.792825. (OA) [SPA] [Ver online]

Abstract: Biomarkers are essential tools for accurate diagnosis and effective prevention, but their validation is a pending challenge that limits their usefulness, even more so with constructs as complex as frailty. Sarcopenia shares multiple mechanisms with frailty which makes it a strong candidate to provide robust frailty biomarkers. Based on this premise, we studied the temporal evolution of cellular interactome in frailty, from independent patients to dependent ones. Overweight is a recognized cause of frailty in aging, so we studied the altered mechanisms in overweight independent elderly and evaluated their aggravation in dependent elderly. This evidence of the evolution of previously altered mechanisms would significantly support their role as real biomarkers of frailty. The results showed a preponderant role of autophagy in interactome control at both different functional points, modulating other essential mechanisms in the cell, such as mitochondrial capacity or oxidative stress. Thus, the overweight provoked in the muscle of the elderly an overload of autophagy that kept cell survival in apparently healthy individuals. This excessive and permanent autophagic effort did not seem to be able to be maintained over time. Indeed, in dependent elderly, the muscle showed a total autophagic inactivity, with devastating effects on the survival of the cell, which

showed clear signs of apoptosis, and reduced functional capacity. The frail elderly are in a situation of weakness that is a precursor of dependence that can still be prevented if detection is early. Hence biomarkers are essential in this context.

15.- DE LA ROZA-DELGADO, B., MARTÍNEZ-FERNÁNDEZ, A., MODROÑO M.S., & ARGAMENTERÍA, A. (2021ª-2022ᵖ). Pradera de *Lolium perenne* y *Trifolium repens* en Asturias. II. Balances nutricionales sobre vacas frisonas en producción a lo largo del año. *ITEA-Información Técnica Económica Agraria,118*(1), 48-68. DOI: 10.12706/itea.2021.039. (OA) [NPF] [Ver online]

Abstract: The temporary ley of *Lolium perenne* and *Trifolium repens* is widely used for milk production in temperate-humid areas. Its nutritional value depends on its chemical composition, ingestibility and subsequent digestive and metabolic use. On the central eastern coast of Asturias, data are available on the evolution of its chemical composition and digestibility. The general objective of this paper was to obtain information on the use of energy and nitrogen ingested. The specific objectives were to determine, throughout the year and for a period of 12 years, the evolution of the energy units, the urinary losses of nitrogen and the recovery of the same in milk, with forage as a single diet or supplemented with different scales and modalities of concentrate. During spring, metabolizable energy at maintenance level (MJ), milk forage units according to INRA 2018 and net lactation energy at feed level 3 (Mcal), per kg of dry matter, ranged respectively between 11.1-11.7; 0.93-1.01 and 1.31-1.49. In summer, they were reduced to 10.2-8.9; 0.87-0.69 and 1.18-0.84. In autumn, spring values gradually recovered. The grass silages had values similar to those of summer grass. Energy supplementation with concentrates significantly elevated metabolicity, feeding level and recovery of raw energy ingested in milk. Nitrogen supplementation significantly increased the recovery of nitrogen ingested in milk and with 15 % crude protein on dry matter, the excretion of nitrogen into the environment was reduced. The evaluated pasture has high potential for milk production in compliance with EU regulations.

16.- DELGADO, A., DAPENA, E., FERNÁNDEZ, E., & LUEDELING, E. (2021). Climatic requirements during dormancy in apple trees from northwestern Spain–Global warming may threaten the cultivation of high-chill cultivars. *European Journal of Agronomy,130*, Art.126374. DOI: 10.1016/j.eja.2021.126374. [CHF] [Ver online]

Abstract: Winter chill is expected to decrease in many mild-winter regions under future climatic conditions. Reliable estimates of the chill requirements (CR) of fruit trees are essential for assessing the current suitability of cultivars and potential climate change impacts on fruit production. We determined chill and heat requirements of ten apple cultivars in northwestern Spain using a bud-forcing method. CR ranged from 59 (‘Granny Smith’) to 90 (‘Regona’) Chill Portions (CP) according to the Dynamic Model. These results indicate that international dessert apple cultivars such as ‘Elstar’ and ‘Granny Smith’ have clearly lower CR than the studied local cultivars. The agro-climatic needs of the traditional apple cultivars are aligned with the historical climate conditions in the region. To assess future apple cultivation in northwestern Spain, we evaluated winter chill availability over the course of the twenty-first century by applying an ensemble of future climate scenarios. Relative to the past, projected winter chill might decline by between 9 and 12 CP under an intermediate global warming scenario and by between 9 and 24 CP under a pessimistic scenario. Despite relatively minor changes, the viability of some local apple cultivars may be jeopardized by their high CR. Results suggest that even a moderate decline in future winter chill, relative to fairly high levels observed in the past, can threaten the economic sustainability of fruit tree orchards composed of high-chill genotypes. Strategies such as growing low- to

moderate-chill cultivars may be critical for sustaining future apple production in the region. Our findings can help guide new breeding strategies aiming to develop climate-resilient cultivars adapted to future environmental conditions.

17.- DELGADO, A., EGEA, J. A., LUEDELING, E., & DAPENA, E. (2021). Agroclimatic requirements and phenological responses to climate change of local apple cultivars in northwestern Spain. *Scientia Horticulturae, 283*, Art.110093. [CHF] [Ver online]

Abstract: In a global warming context, analyses of historic temperature records are essential to understand the potential impacts of climate change on spring phenology. To estimate flowering trends over recent decades, we analyzed long-term temperature and phenology records of eleven local apple cultivars in Asturias (northwestern Spain) in a temperate oceanic climate. Our results show that, over a period of 30 years, bloom dates of the local cultivars have experienced relatively minor changes, considering that temperatures increased strongly since 1978, by 0.30 °C per decade. An explanation for this weak phenological response to warming may be that these temperature changes only had a small effect on overall chill accumulation, but possibly delayed the onset date of endodormancy, which may have counteracted phenology-advancing effects of warming in spring. At present, chill accumulation in this area is high, at an average of 96 Chill Portions from November to March, which indicates that chill is not currently a limiting factor for the quality of flowering and fruiting in the study area. We used Partial Least Squares (PLS) regression to delineate an effective chilling period between November 12th and February 9th and effective heat accumulation between March 15th and May 4th. While these periods appear plausible, we noticed that this approach was unable to identify well-known differences in chilling requirements among many of the cultivars, with similar chill needs determined for many of them. This observation may be explained by inaccurate expectations about cultivars' climatic needs, by inaccuracy of the chill (and possibly heat) model or, most concerning, by inability of the PLS approach to correctly identify the chilling periods of apple cultivars in this region. Bloom dates were similarly responsive to mean temperature during the chill and the heat accumulation phases, indicating that both processes need to be considered when predicting future phenology.

18.- DELGADO, A., QUINET, M., & DAPENA, E. (2021). Analysis of the variability of floral and pollen traits in apple cultivars—selecting suitable pollen donors for cider apple orchards. *Agronomi-Basel, 11*(9), Art.1717. DOI: 10.3390/agronomy11091717. (OA) [CHF] [Ver online]

Abstract: Most apple trees (*Malus domestica* Borkh.) are self-incompatible and fruit yield depends on cross-pollination between genetically compatible cultivars with synchronous flowering. Flowering intensity can vary strongly among years due to the biennial bearing habit of the cultivars. The knowledge of the phenological stages and floral and pollen characteristics is essential to select suitable pollen donors. We evaluated the phenotypic variability of flowering-related traits (i.e., flowering phenology, flowering intensity, pollen production and pollen quality) in 45 apple cultivars over two successive flowering seasons. Large phenotypic variability was found among the studied cultivars indicating that the local germplasm collection provides a good source of genetic and phenotypic diversity. However, low correlations were observed between floral biology traits and, consequently, the improvement in one trait seems not to affect other traits. Some of the cultivars such as 'Perurico' and 'Raxila Dulce' regularly produced copious amounts of high-quality pollen which can improve the pollen load dispersion leading to a most effective pollination process. We did not identify statistically significant correlations between pollen attributes and the biennial bearing phenomenon. The large variation in bloom dates from year-to-year observed under a typical Oceanic climate makes it advisable to combine cultivars in new plantings.

19.- DENOYELLE, L., TALOUARN, E., BARDOU, P., DENOYELLE, L., TALOUARN, E., BARDOU, P., COLLI, L., ALBERTI, A., DANCHIN, C., DEL CORVO, M., ENGELEN, S., ORVAIN, C., PALHIÈRE, I., RUPP, R., SARRY, J., SALAVATI, M., AMILLS, M., CLARK, E., CREPALDI, P., FARAUT, T., MASIGA, C.W., POMPANON, F., ROSEN, B.D., STELLA, A., VAN TASSELL, C.P., TOSSER-KLOPP, G., & THE VARGOATS CONSORTIUM [KIJAS, J., GULDBRANDTSEN, B., KANTANEN, J., DUBY, D., MARTIN, P., DANCHIN, C., ALLAIN, D., ARQUET, R., MANDONNET, N., NAVES, M., PALHIÈRE, I., RUPP, R., POMPANON, F., REZAEI, H.R., CAROLAN, S., FORAN, M., STELLA, A., AJMONE-MARSAN, P., COLLI, L., CRISÀ, A., MARLETTA, D., CREPALDI, P., OTTINO, M., RANDI, E., BENJELLOUN, B., LENSTRA, H., MOAEEN-UD-DIN, M., REECY, J., GOYACHE, F., ÁLVAREZ, I., AMILLS, M., SÀNCHEZ, A., CAPOTE, J., JORDANA, J., PONS, A., MARTÍNEZ, A., MOLINA, A., ROSEN, B., VISSER, C., DRÖGEMÜLLER, C., LUIKART,G ., MASIGA, C.W., MUJIBI, D.F., MRUTTU, H.A., GONDWE, T., SIKOSANA, J., TAELA, M.G.,& NASH, O.] (2021). VarGoats project: a dataset of 1159 whole-genome sequences to dissect *Capra hircus* global diversity. *Genetics Selection Evolution 53*, Art. 86. DOI: 10.1186/s12711-021-00659-6. (OA) [GRA] [Ver online]

Abstract: Background:Since their domestication 10,500 years ago, goat populations with distinctive genetic backgrounds have adapted to a broad variety of environments and breeding conditions. The VarGoats project is an international 1000-genome resequencing program designed to understand the consequences of domestication and breeding on the genetic diversity of domestic goats and to elucidate how speciation and hybridization have modeled the genomes of a set of species representative of the genus *Capra*.
Findings:A dataset comprising 652 sequenced goats and 507 public goat sequences, including 35 animals representing eight wild species, has been collected worldwide. We identified 74,274,427 single nucleotide polymorphisms (SNPs) and 13,607,850 insertion-deletions (InDels) by aligning these sequences to the latest version of the goat reference genome (ARS1). A Neighbor-joining tree based on Reynolds genetic distances showed that goats from Africa, Asia and Europe tend to group into independent clusters. Because goat breeds from Oceania and Caribbean (Creole) all derive from imported animals, they are distributed along the tree according to their ancestral geographic origin.
Conclusions:We report on an unprecedented international effort to characterize the genome-wide diversity of domestic goats. This large range of sequenced individuals represents a unique opportunity to ascertain how the demographic and selection processes associated with post-domestication history have shaped the diversity of this species. Data generated for the project will also be extremely useful to identify deleterious mutations and polymorphisms with causal effects on complex traits, and thus will contribute to new knowledge that could be used in genomic prediction and genome-wide association studies.

20.- DÍAZ, M., CONCEPCIÓN, E.D., MORALES, M.B., ALONSO, J.C., AZCÁRATE, F.M., BARTOMEUS, I., BOTA, G., BROTONS, L., GARCÍA,

D., GIRALT, D., GUTIÉRREZ, J.E., LÓPEZ-BAO, J.V., MAÑOSA, S., MILLA, R., MIÑARRO, M., NAVARRO, A., OLEA, P.P., PALACÍN, C., PECO, B., REY, P.J., SEOANE, J., SUÁREZ-SEOANE, S., SCHÖB, C., TARJUELO, R., TRABA, J., VALERA, F., & VELADO-ALONSO, E. (2021). Environmental objectives of Spanish agriculture: scientific guidelines for their effective implementation under the common agricultural policy 2023-2030. *Ardeola 68*(2), 445-460. DOI: 10.13157/arla.68.2.2021.fo1. (OA) [CHF] [Ver online]

Abstract: The next reform of the EU Common Agricultural Policy (CAP) for the period 2021-2027 (currently extended to 2023-2030) requires the approval by the European Commission of a Strategic Plan with environmental objectives for each Member State. Here we use the best available scientific evidence on the relationships between agricultural practices and biodiversity to delineate specific recommendations for the development of the Spanish Strategic Plan. Scientific evidence shows that Spain should (1) identify clear regional biodiversity targets and the landscape-level measures needed to achieve them; (2) define ambitious and complementary criteria across the three environmental instruments (enhanced conditionality, eco-schemes, and agri-environmental and climate measures) of the CAP's Green Architecture, especially in simple and complex landscapes; (3) ensure that other CAP instruments (areas of nature constraints, organic farming and protection of endangered livestock breeds and crop varieties) really support biodiversity; (4) improve farmers' knowledge and adjust measures to real world constraints; and (5) invest in biodiversity and ecosystem service monitoring in order to evaluate how the Plan achieves regional and national targets and to improve measures if targets are not met. We conclude that direct assessments of environmental objectives are technically and economically feasible, can be attractive to farmers, and are socially fair and of great interest for improving the environmental effectiveness of CAP measures. The explicit and rigorous association of assessments and monitoring, relating specific environmental indicators to regional objectives, should be the main criterion for the approval of the Strategic Plan in an environmentally-focused CAP 2023-2030.—Díaz, M. *et al*. (2021). Environmental objectives of Spanish agriculture: scientific guidelines for their effective implementation under the Common Agricultural Policy 2023-2030.

21.- ESPÍ, A., DEL CERRO, A., OLEAGA, Á., RODRÍGUEZ-PÉREZ, M., LÓPEZ, C. M., HURTADO, A., RODRÍGUEZ-MARTÍNEZ, L. D., BARANDIKA, J.F., & GARCÍA-PÉREZ, A. L. (2021). One health approach: An overview of Q Fever in LIvestock, wildlife and humans in Asturias (Northwestern Spain). *Animals, 11*(5), Art. 1395. DOI: 10.3390/ani11051395. (OA) [SA] [Ver online]

Abstract: This study aimed to investigate the seroprevalence of *C. burnetii* in domestic ruminants, wild ungulates, as well as the current situation of Q fever in humans in a small region in northwestern Spain where a close contact at the wildlife–livestock–human interface exists, and information on *C. burnetii* infection is scarce. Seroprevalence of *C. burnetii* was 8.4% in sheep, 18.4% in cattle, and 24.4% in goats. Real-time PCR analysis of environmental samples collected in 25 livestock farms detected *Coxiella* DNA in dust and/or aerosols collected in 20 of them. Analysis of sera from 327 wild ungulates revealed lower seroprevalence than that found in domestic ruminants, with 8.4% of Iberian red deer, 7.3% chamois, 6.9% fallow deer, 5.5% European wild boar and 3.5% of roe deer harboring antibodies to *C. burnetii*. Exposure to the pathogen in humans was determined by IFAT analysis of 1312 blood samples collected from patients admitted at healthcare centers with Q fever compatible symptoms, such as fever and/or pneumonia. Results showed that 15.9% of the patients had IFAT titers ≥ 1/128 suggestive of probable acute infection. This study is an example of a One Health approach with medical and veterinary institutions involved in investigating zoonotic diseases.

22.- FERNÁNDEZ, H., GROSSMANN, J., GAGLIARDINI, V., FEITO, I., RIVERA, A., RODRIGUEZ, L., QUINTANILLA, L., QUESADA, V., CANAL M.J. & GROSSNIKLAUS, U. (2021). Sexual and Apogamous species of woodferns show different protein and phytohormone profiles. *Frontiers in Plant Science, 12*, Art. 718932. DOI: 10.3389/fpls.2021.718932. (OA) [CHF] [Ver online]

Abstract: The gametophyte of ferns reproduces either by sexual or asexual means. In the latter, apogamy represents a peculiar case of apomixis, in which an embryo is formed from somatic cells. A proteomic and physiological approach was applied to the apogamous fern *Dryopteris affinis* ssp. *affinis* and its sexual relative *D. oreades*. The proteomic analysis compared apogamous vs. female gametophytes, whereas the phytohormone study included, in addition to females, three apogamous stages (filamentous, spatulate, and cordate). The proteomic profiles revealed a total of 879 proteins and, after annotation, different regulation was found in 206 proteins of *D. affinis* and 166 of its sexual counterpart. The proteins upregulated in *D. affinis* are mostly associated to protein metabolism (including folding, transport, and proteolysis), ribosome biogenesis, gene expression and translation, while in the sexual counterpart, they account largely for starch and sucrose metabolism, generation of energy and photosynthesis. Likewise, ultra-performance liquid chromatography-tandem spectrometry (UHPLC-MS/MS) was used to assess the levels of indol-3-acetic acid (IAA); the cytokinins: 6-benzylaminopurine (BA), trans-Zeatine (Z), trans-Zeatin riboside (ZR), dyhidrozeatine (DHZ), dyhidrozeatin riboside (DHZR), isopentenyl adenine (iP), isopentenyl adenosine (iPR), abscisic acid (ABA), the gibberellins GA_3 and GA_4, salicylic acid (SA), and the brassinosteroids: brassinolide (BL) and castasterone (CS). IAA, the cytokinins Z, ZR, iPR, the gibberellin GA_4, the brassinosteoids castasterone, and ABA accumulated more in the sexual gametophyte than in the apogamous one. When comparing the three apogamous stages, BA and SA peaked in filamentous, GA_3 and BL in spatulate and DHRZ in cordate gametophytes. The results point to the existence of large metabolic differences between apogamous and sexual gametophytes, and invite to consider the fern gametophyte as a good experimental system to deepen our understanding of plant reproduction.

23.- FERNÁNDEZ, M., PÉREZ, V., FUERTES, M., BENAVIDES, J., ESPINOSA, J., MENÉNDEZ, J., GARCÍA-PÉREZ, A. L., & FERRERAS, M. C. (2021). Pathological study of facial eczema (*Pithomycotoxicosis*) in sheep. *Animals, 11*(4), Art. 1070. DOI: 10.3390/ani11041070. (OA) [SPA] [Ver online]

Abstract: Facial eczema (FE) is a secondary photosensitization disease of farm ruminants caused by the sporidesmin A, produced in the spores of the saprophytic fungus *Pithomyces chartarum*. This study communicates an outbreak of ovine FE in Asturias (Spain) and characterizes the serum biochemical pattern and the immune response that may contribute to liver damage, favoring cholestasis and the progression to fibrosis and cirrhosis. Animals showed clinical signs of photosensitivity, with decrease of daily weight gain and loss of wool and crusting for at least 6 months after the FE outbreak. Serum activity of γ-glutamyltransferase and alkaline phosphatase were significantly increased in sheep with skin lesions. In the acute phase, edematous skin lesions in the head, hepatocytic and canalicular cholestasis in centrilobular regions, presence of neutrophils in small clumps surrounding deposits of bile pigment, ductular

proliferation, as well as cholemic nephrosis, were observed. Macrophages, stained positively for MAC387, were found in areas of canalicular cholestasis. In the chronic phase, areas of alopecia and crusting were seen in the head, and the liver was atrophic with large regeneration nodules and gallstones. Fibrosis around dilated bile ducts, "typical" and "atypical" ductular reaction and an inflammatory infiltrate composed of lymphocytes and pigmented macrophages, with iron deposits and lipofuscin, were found. The surviving parenchyma persisted with a jigsaw pattern characteristic of biliary cirrhosis. Concentric and eccentric myointimal proliferation was found in arteries near damaged bile ducts. In cirrhotic livers, stellated cells, ductular reaction, ectatic bile ducts and presence of M2 macrophages and lymphocytes, were observed in areas of bile ductular reaction.

24.- FUENTE-GARCÍA, C., ALDAI, N., SENTANDREU, E., OLIVÁN, M., FRANCO, D., GARCÍA-TORRES, S., BARRON, L. J. R., & SENTANDREU, M. A. (2021). Caspase activity in post mortem muscle and its relation to cattle handling practices. *Journal of the Science of Food and Agriculture, 101*(15), 6258-6264. DOI: 10.1002/jsfa.11293. (OA) [SPA] [Ver online]

Abstract: Background: Animal handling practices are one of the factors majorly affecting animal metabolism prior to slaughter. This phenomenon increases the occurrence of meat quality defects such as dark cutting-beef, causing high economical losses in the meat industry. Under this framework, the assessment of apoptosis onset in *post mortem* muscle was proposed as a novel approach to reveal biochemical characteristics in several Spanish bovine breeds (Asturiana de los Valles, Retinta and Rubia Gallega) managed under different production systems (intensive *versus* semi-extensive) and transport/lairage conditions (mixing *versus* not mixing with unfamiliar animals). To do so, the activities of initiator caspase 9 and executioner caspases 3/7 were determined in *Longissimus thoracis et lumborum* muscle at three early *post mortem* times (2, 8, and 24 h).
Results: Breed effect and transport/lairage conditions were the most relevant factors that influenced both caspase activities over *post mortem* time, showing Rubia Gallega breed a completely different behavior compared to Asturiana de los Valles and Retinta breeds. Moreover, it is postulated that apoptosis cascade is initiated via the activation of caspase 9 under hypoxic or metabolic stress followed by the activation of executioner caspases 3/7.
Conclusions: Assessment of apoptosis on *post mortem* muscle can be a novel approach to study the influence of animal handling on muscle metabolism and *post mortem* cell death and its consequences on meat quality traits**.**

25.- FUENTE-GARCÍA, C., SENTANDREU, M. A., ALDAI, N., OLIVÁN, M., & SENTANDREU, E. (2021). Proteomic pipeline for biomarker hunting of defective bovine meat assisted by liquid chromatography-mass spectrometry analysis and chemometrics. *Journal of Proteomics, 238*, Art.104153. [SPA] [Ver online]

Abstract: A wide variety of factors prior to slaughter may affect the stress status of beef cattle, giving rise to well-known 'dark-cutting' defective meats characterised by a high ultimate pH (pHu). To understand the underlying mechanisms of pHu fluctuations in beef cattle there was studied the proteome changes caused by pre-slaughter stress through a gel-free proteomic approach. Comparative peptidomic analysis was carried out on 12 loin samples at 24 h post-mortem from *Longissimus thoracis et lumborum* bovine muscle of crossbred animals, previously sorted into two different groups according to their pHu values: normal (pHu < 6.0) and high (pHu ≥ 6.0). Tryptic peptides from direct protein extracts were approached by combining untargeted (intact

mass, MS^1) and targeted (Selected Reaction Monitoring, SRM) quantitative LC-MS assays followed by chemometric analysis. Seventeen peptide biomarkers belonging to 10 different proteins appropriately discriminated sample groups assayed. Results may promote the use of this simple and effective methodology towards the creation of new insights in meat quality research.

26.- GARCÍA-FERNÁNDEZ, C., CAMPA, A., SOLER GARZÓN, A., MIKLAS, P. & FERREIRA, J., J. (2021). GWAS of pod morphological and color characters in common bean*. BMC Plant Biology 21*, Art.184. DOI: 10.1186/s12870-021-02967-x. (OA) [CHF] [Ver online]

Abstract: Background: Common bean (*Phaseolus vulgaris* L.) is an important legume species which can be consumed as immature pods and dry seeds after re-hydration and cooking. Many genes and QTL, and epistatic interactions among them, condition pod morphological traits. However, not all them have been mapped or validated nor candidate genes proposed. We sought to investigate the genomic regions conditioning pod morphological and color characters through GWAS.

Results: Single and multi-locus genome wide association analysis was used to investigate pod traits for a set of 301 bean lines of the Spanish Diversity Panel (SDP). The SDP was genotyped with 32,812 SNPs obtained from Genotyping by Sequencing. The panel was grown in two seasons and phenotypic data were recorded for 17 fresh pods traits grouped in four pod characters: pod length, pod cross-section, pod color, and number of seeds per pod. In all, 23 QTL for pod length, 6 for cross-section, 18 for pod color, 6 for number of seeds per pod and 9 associated to two or more pod characters were detected. Most QTL were located in the telomeric region of chromosomes Pv01, Pv02, Pv04, Pv08, Pv09 and Pv10. Eighteen detected QTL co-localized with 28 previously reported QTL. Twenty-one potential candidate genes involving developmental processes were detected underlying 11 QTL for pod morphological characters, four of them homologous to *A. thaliana* genes *FIS*2, *SPL10*, *TTG2* and *AML4* affecting silique size. Eight potential candidate genes involved in pigment synthesis, were found underlying five QTL for pod color.

Conclusions: GWAS for pod morphological and color characters in the bean Spanish Diversity Panel revealed 62 QTL, 18 co-localized with previously reported QTL, and 16 QTL were underlain by 25 candidate genes. Overall 44 new QTL identified and 18 existing QTL contribute to a better understanding of the complex inheritance of pod size and color traits in common bean and open the opportunity for future validation works.

27.- GARCÍA-FERNÁNDEZ, C., CAMPA, A., & FERREIRA, J. J. (2021). Dissecting the genetic control of seed coat color in a RIL population of common bean (*Phaseolus vulgaris L*.). *Theoretical and Applied Genetics*, *134*, 3687-3698 .DOI: 10.1007/s00122-021-03922-y. [CHF] [Ver online]

Abstract: Seed coat color is an important characteristic of common bean (*Phaseolus vulgaris* L.) associated with the marketability of dry bean cultivars, quality and nutritional characteristics of seed, as well as response to pathogens. In this study, the genetic control of seed coat color in a recombinant inbred line population (175 lines) obtained from the cross 'TU' × 'Musica' was investigated. Phenotypic segregation fitted 1:1 for white vs. nonwhite, and 3:1 for brown versus black, indicating the involvement of three independent genes, one controlling white color and two (with epistatic interaction) controlling black color. Using a genetic map built with 842 SNPs, the gene responsible for the white seed coat was mapped on the linkage group Pv07, in the position previously described for the *P* gene. For the black seed coat phenotype, two genes were mapped to the beginning of chromosomes Pv06 and Pv08, in the positions estimated for the *V* gene and the complex *C* locus, respectively, by classical studies. The involvement of these two genomic regions was verified through two crosses between three selected RILs exhibiting complementary and dominant inheritance, in which the TU alleles for both genes resulted in a black phenotype. Two genes involved in the anthocyanin biosynthesis pathway were proposed as candidate genes*:* *Phvul.006G018800* encoding a flavonoid 3′5’hydroxylase and *Phvul.008G038400* encoding MYB113 transcription factor. These findings add knowledge to the complex network of genes controlling seed coat color in common bean as well as providing genetic markers to be used in future genetic analysis or plant breeding.

28.-GARCÍA-SÁNCHEZ, M., JIMÉNEZ-PELAYO, L., VÁZQUEZ, P., HORCAJO, P., REGIDOR-CERRILLO, J., JIMÉNEZ-MELÉNDEZ, A., OSORO, K., ORTEGA MORA, L.M., & COLLANTES-FERNÁNDEZ, E. (2021). Maternal and foetal cellular immune responses in dams infected with high-and low-virulence isolates of *Neospora caninum* at mid-gestation. *Frontiers in Cellular and Infection Microbiology, 11*, Art. 684670. DOI: 10.3389/fcimb.2021.684670. (OA) [SPA] [Ver online]

Abstract: Bovine neosporosis is currently considered one of the main causes of abortion in cattle worldwide and the outcome of the infection is, in part, determined by *Neospora caninum* isolate virulence. However, the dam and foetal immune responses associated with this factor are largely unknown. We used a model of bovine infection at day 110 of gestation to study the early infection dynamics (10- and 20-days post-infection, dpi) after experimental challenge with high- and low-virulence isolates of *N. caninum* (Nc-Spain7 and Nc-Spain1H, respectively). In the present work, dam peripheral cellular immune responses were monitored twice a week from -1 to 20 dpi. At different time points, IFN-γ and IL-4 production was investigated in stimulated dam blood and the percentage of monocytes, NK cells, B cells and T cells (CD4+, CD8+ and γδ) in peripheral blood mononuclear cells (PBMC) were determined by flow cytometry. In addition, maternal iliofemoral lymph nodes and foetal spleen and thymus were collected at 10 and 20 dpi for the study of the same cell subpopulations. Peripheral immune response dynamics were similar after the infection with both isolates, with a significant increase in the percentage of CD4+ T cells at 6 and 9 dpi in PBMC, coincident with the higher levels of IFN-γ and IL-4 release. However, the levels of IFN-γ were significantly higher and an increase in CD8+ T cells at 9, 13 and 20 dpi was observed in the dams infected with Nc-Spain7. Nc-Spain1H infection induced higher IL4 levels in stimulated blood and a higher CD4+/CD8+ ratio in PBMC. The analysis of the maternal iliofemoral lymph node showed a significant enhancement in the percentage of NK, CD4+ and CD8+ T cells for the animals infected with the highly virulent isolate and euthanized at 20 dpi. Regarding the foetal responses, the most remarkable result was an increase in the percentage of monocytes at 20 dpi in the spleen of foetuses from both infected groups, which suggests that foetuses were able to respond to *N. caninum* infection at mid gestation. This work provides insights into how isolate virulence affects the maternal and foetal immune responses generated against *N. caninum*, which may influence the course of infection.

29.- GARCÍA-TORRES, S., CABEZA DE VACA, M., TEJERINA, D., ROMERO-FERNÁNDEZ. M.P., ORTI, A., FRANCO, D., SENTANDREU, M.A., & OLIVÁN, M. (2021). Assessment of stress by serum biomarkers in calves and their relationship to ultimate pH as an indicator of meat quality. *Animals, 11*(8), Art. 2291. DOI: 10.3390/ani1108229. (OA) [SPA] [Ver online]

Abstract: Seventy-eight calves from Asturiana de los Valles, Retinta, and Rubia Gallega breeds, under extensive and intensive farm systems and animal mixing and non-mixing conditions, and during the transport and lairage in slaughterhouses, were studied. This research aimed to study the effect of breed, farm system and mixing conditions on serum biomarkers (cortisol, lactate, glucose, serum amyloid A, haptoglobin, and C-reactive protein) and their relationship with pH_u at slaughter time, and to evaluate the response of the serum biomarkers of calves throughout fattening period. Moreover, this study aims to evaluate the response of the biomarkers in each breed during the fattening period. At slaughter time, cortisol and lactate were affected by BreedxFarm; Retinta showed the opposite pattern to the others and revealed the highest glucose in extensive farm systems. Rubia Gallega in mixing revealed the highest Amyloid A and haptoglobin. Extensive calves in mixing conditions showed the highest glucose. There was a relationship among the variables cortisol, lactate, Amyloid A, and pH_u. Slaughter time was a major stressor, and the stress response was mainly affected by breed. At slaughter, several biomarkers should be considered.

30.- GARRATT, M.P.D., DE GROOT, A. G., ALBRECHT, M., BOSCH, J., BREEZE, T. D., FOUNTAIN, M., KLEIN, A. MCKERCHAR, M., PARK, M., POTTS, S. G.1 PUFAL, G., RADER, R., SENAPATHI, G. D.1, ANDERSSON, G. K. S., BERNAUER, O. M., BLITZER, E. J., BOREUX, V., CAMPBELL, A., FOLDESI, R., GARCÍA, D., GARIBALDI, L., HAMBÄCK, P. A., KIRKITADZE, G., KOVÁCS-HOSTYÁNSZKI, A., MARTINS, K. T., MCKENDRICK, L., MIÑARRO, M., RADZEVICIUTE, R., ROQUER, L., SAMNEGÅRD, U., VEREECKEN, N. J. WEBBER, S.1, JAPOSHVILI, G. & ZHUSUPBAEVA, A. (2021). Opportunities to reduce pollination deficits and production shortfalls in an important insect pollinated crop. *Ecological Applications*, *31*(8), Art. e02445. DOI: 10.1002/eap.2445. (OA) [CHF] [Ver online]

Abstract: Pollinators face multiple pressures and there is evidence of populations in decline. As demand for insect-pollinated crops increases, crop production is threatened by shortfalls in pollination services. Understanding the extent of current yield deficits due to pollination and identifying opportunities to protect or improve crop yield and quality through pollination management is therefore of international importance. To explore the extent of "pollination deficits," where maximum yield is not being achieved due to insufficient pollination, we used an extensive dataset on a globally important crop, apples. We quantified how these deficits vary between orchards and countries and we compared "pollinator dependence" across different apple varieties. We found evidence of pollination deficits and, in some cases, risks of overpollination were even apparent for which fruit quality could be reduced by too much pollination. In almost all regions studied we found some orchards performing significantly better than others in terms of avoiding a pollination deficit and crop yield shortfalls due to suboptimal pollination. This represents an opportunity to improve production through better pollinator and crop management. Our findings also demonstrated that pollinator dependence varies considerably between apple varieties in terms of fruit number and fruit quality. We propose that assessments of pollination service and deficits in crops can be used to quantify supply and demand for pollinators and help to target local management to address deficits although crop variety has a strong influence on the role of pollinators.

31.- GIMENO, I., GARCÍA-MANRIQUE, P., CARROCERA, S., LÓPEZ-HIDALGO, C., VALLEDOR, L., MARTÍN-GONZÁLEZ, D., & GÓMEZ, E.

(2021). The metabolic signature of in vitro produced bovine embryos helps predict pregnancy and birth after embryo transfer. *Metabolites, 11*(8), Art. 484. DOI: 10.3390/metabo11080484. [GRA] [Ver online]

Abstract: In vitro produced (IVP) embryos show large metabolic variability induced by breed, culture conditions, embryonic stage and sex and gamete donors. We hypothesized that the birth potential could be accurately predicted by UHPLC-MS/MS in culture medium (CM) with the discrimination of factors inducing metabolic variation. Day-6 embryos were developed in single CM (modified synthetic oviduct fluid) for 24 h and transferred to recipients as fresh (28 ETs) or frozen/thawed (58 ETs) Day-7 blastocysts. Variability was induced with seven bulls, slaughterhouse oocyte donors, culture conditions (serum + Bovine Serum Albumin [BSA] or BSA alone) prior to single culture embryonic stage records (Day-6: morula, early blastocyst, blastocyst; Day-7: expanding blastocyst; fully expanded blastocysts) and cryopreservation. Retained metabolite signals (6111) were analyzed as a function of pregnancy at Day-40, Day-62 and birth in a combinatorial block study with all fixed factors. We identified 34 accumulated metabolites through 511 blocks, 198 for birth, 166 for Day-62 and 147 for Day-40. The relative abundance of metabolites was higher within blocks from non-pregnant (460) than from pregnant (51) embryos. Taxonomy classified lipids (12 fatty acids and derivatives; 224 blocks), amino acids (12) and derivatives (3) (186 blocks), benzenoids (4; 58 blocks), tri-carboxylic acids (2; 41 blocks) and 5-Hydroxy-l-tryptophan (2 blocks). Some metabolites were effective as single biomarkers in 95 blocks (Receiver Operating Characteristic – Area Under the Curve [ROC-AUC]: 0.700–1.000). In contrast, more accurate predictions within the largest data sets were obtained with combinations of 2, 3 and 4 single metabolites in 206 blocks (ROC-AUC = 0.800–1.000). Pregnancy-prone embryos consumed more amino acids and citric acid, and depleted less lipids and cis-aconitic acid. Big metabolic differences between embryos support efficient pregnancy and birth prediction when analyzed in discriminant conditions.

32.- GIMENO I., GARCÍA-MANRIQUE ,P., CARROCERA S., LÓPEZ-HIDALGO C., VALLEDOR L., MARTÍN-GONZÁLEZ D., GÓMEZ, E. (2021). Noninvasive prediction of pregnancy and birth in cattle by liquid chromatography-high-resolution mass spectrometry analysis of embryo culture medium. *Reproduction, Fertility and Developmen,t 34*(2), 266. DOI: 10.1071/RDv34n2Ab61. (OA) [GRA] [Ver online]

Abstract: Estimation of the embryo competence to reach birth is a major objective in reproductive technology. Discrimination of the metabolic variation induced by fixed factors (FF) (i.e. culture conditions, embryonic stage, bull breed, and embryo cryopreservation) may help refine estimations. We hypothesised that pregnancy and birth of transferred fresh and frozen IVP bovine embryos can be predicted by liquid chromatography-high-resolution mass spectrometry (LC-HRMS) analysis of culture medium (CM). Oocytes fertilised and cultured in groups in modified synthetic oviductal fluid with bovine serum albumin (SOFaaci-BSA) with or without fetal calf serum (FCS), were later individually cultured in protein-free CM for 24h, and transferred to recipients as fresh (28 ETs; 57% birth rate (BR)) or frozen/thawed (58 ETs; BR Day 7 embryos: 46%; BR Day 8 embryos: 12.5% (1/8)) blastocysts. Spent CM was analysed by LC-HRMS. Chromatograms were processed (MZmine v2.53 and R package pRocessomics) to retain 6111 signals, which were analysed as a function of pregnancy at Day 40, Day 62, and birth by sparse partial least squares-discriminant analysis (PLS-DA) and orthogonal PLS-DA. Both within the entire dataset and in sample blocks with any single FF, DA did not yield sample separation. However, some blocks of samples formed by two FFs differed by permutation (e.g. a block of $N = 48$ frozen (FF1) embryos cultured with BSA (FF2) at birth: Q^2: $P < 0.03$, R^2Y: $P < 0.01$). Thereafter, a univariate study analysed all combinations of FF in blocks under the criteria of fold change (FCh) $> |2|$ (pregnancy/non-pregnancy) and $P < 0.05$ or tendencies $0.05 > P < 0.10$ (ANOVA and Kruskal–Wallis test) and area under the receiver operator characteristic curve (ROC-AUC) >0.700 with FCh $> |2|$) and *t*-test $P < 0.05$ and $P < 0.10$. At this stage, 34 significantly accumulated metabolites were identified through 198 blocks for birth, 166 for Day 62 and 147 for Day 40. Accumulated metabolites were higher in blocks from non-pregnancy-prone (460) than from pregnancy-prone (51) embryos. Blocks of embryos transferred frozen accounted for 33 metabolites (with 16 of them not represented in fresh embryos); 17 metabolites were independent of embryo cryopreservation; and only one metabolite was exclusive of fresh embryos (5-hydroxy-L-Trp). Classes were amino acids (12) and derivatives (3) (186 blocks), lipids (12; 224 blocks), benzenoids (4; 58 blocks), tri-carboxylic acids (2; 41 blocks) and 5-hydroxy-L-Trp (2 blocks). Some metabolites were predictive as single biomarkers in 95 blocks (ROC-AUC: 0.700–1.000), but more accurate predictions in the largest datasets were given by combinations of 2, 3, and 4 single metabolites in 206 blocks (ROC-AUC = 0.800–1.000). Dimethyl adypate had more blocks on Day 40 (18) and Day 62 (19) than at birth (4), suggesting a candidate to predict late miscarriage. Generally, pregnancy-prone embryos depleted less lipids and *cis*-aconitic acid, and consumed more amino acids and citric acid. We suggest that competent embryos accumulate fewer lipids in their cells. Our work is the first to identify high numbers of birth biomarkers by LC-HRMS in bovine embryos. Biomarker discovery can be improved upon discrimination of factors that induce metabolic variability in embryos.

33.- GIMENO I., GARCÍA-MANRIQUE P., GATIEN J., CARROCERA S., GOYACHE F., BERDASCO M., SALVETTI P., GÓMEZ E. (2021). Early metabolic reading of calf fitness in the embryo and the recipient. *Reproduction, Fertility and Development, 34*(2), 315. DOI: 10.1071/RDv34n2Ab154. (OA) [GRA] [Ver online]

Abstract: Early in development, metabolites in the embryo culture medium (CM) and recipient blood may influence birth to term; thus, traits of the calf may be retrospectively associated with the embryo and recipient metabolome. We investigated such metabolomic contributions to calf fitness. Oocytes were *in vitro* fertilised and embryos cultured in synthetic oviduct fluid with amino acids, citrate, and myo-inositol (SOFaaci) in groups with and without fetal calf serum until Day−6, and then singly for 24 h without protein. Calves were born from fresh ($N = 12$) and frozen ($N = 23$) transferred embryos. Metabolites were previously identified in CM by ultra-high performance liquid chromatography with tandem mass spectroscopy (UHPLC-MS/MS; $N = 37$), and in recipient plasma by ^{1}H-nuclear magnetic resonance ($N = 35$). Thirteen calf clinical and morphological signs and 18 analytes (Vetscan i-STAT One analyser) were measured at birth, before and after colostrum intake. Feature values from calf, recipient, and CM were processed together by principal component (PC) analysis to identify covariates. The effects modulating each PC (colostrum intake; embryo freezing; FCS in culture; recipient, bull and calf breed; calf sex and embryonic stage) were calculated with a GLM model. Log-transformed covariates were analysed in debiased sparse partial correlation networks within each PC. Thereafter, metabolic pathways (KEGG) were identified in each PC for CM, recipient, and calf. Four PCs explained 14.19, 9.86, 7.96, and 6.03% of variance. PC1 showed 27 recipient hits (>25 pathways) clustered with mother weight at birth and only two hits in CM (L-Val and Lauroyl diethanolamide). PC2 associated calf (Nitrogen metabolism ($P < 0.004$); Gly, Ser and Thr metabolism [$P < 0.021$]) and CM pathways (Phe, Tyr and Trp biosynthesis; Phe metabolism; glutathione (GSH) metabolism ($P = 0.003$ to 0.053); and Aminoacyl-tRNA (aa-tRNA) biosynthesis), with only pyruvate and lactate up-regulated in recipients. PC3 linked calf gestation, growth and redox status, with CM (Citrate cycle; Glyoxylate and dicarboxylate metabolism; and Ala, Asp and Glu metabolism ($P = 0.0001$ to 0.036)) and recipient pathways (Gly, Ser and Thr metabolism; GSH metabolism; Phe metabolism; Biotin metabolism; aa-tRNA

biosynthesis; Val, Leu, and Ile biosynthesis; and Arg ($P = 0.0001$ to 0.053) biosynthesis). PC4 did not show patterns among calf Na^+, Cl^-, sO_2 and TO_2 (up-regulated) and nasal flux (down-regulated), but they covariated with CM (aa-tRNA biosynthesis; Val, Leu, and Ile biosynthesis; Val, Leu, and Ile degradation; Lys degradation; GSH metabolism; Arg and Pro metabolism; Gln and Glu metabolism; Nitrogen metabolism ($P = 1.95E-09$ to 0.0389); Phe, Tyr and Trp biosynthesis; and Glyoxylate and dicarboxylate metabolism) and recipient pathways (Val, Leu and Ile biosynthesis; aa-tRNA biosynthesis; and His metabolism ($P = 6.97E-05$ to 0.031)). PC3 was affected by embryo freezing ($P < 0.006$), culture ($P = 0.085$) and Day-6 stage ($P < 0.003$), while PC1, PC2 and PC4 only showed breed effects. Culture conditions during early development may entail long-term effects on the offspring. Regulated pathways in the embryo can be explored in association with calf fitness.

34.- GÓMEZ, E., CANELA, N., HERRERO, P., CERETO, A., GIMENO, I., CARROCERA, S., MARTÍN-GONZÁLEZ, D., MURILLO, A., & MUÑOZ, M. (2021). Metabolites secreted by bovine embryos in vitro predict pregnancies that the recipient plasma metabolome cannot, and vice versa. *Metabolites, 11*(3), Art. 162. (OA) [GRA] [Ver online]

Abstract: This work describes the use of mass spectrometry-based metabolomics as a non-invasive approach to accurately predict birth prior to embryo transfer (ET) starting from embryo culture media and plasma recipient. Metabolomics was used here as a predictive platform. Day-6 in vitro produced embryos developed singly in modified synthetic oviduct fluid culture medium (CM) drops for 24 h were vitrified as Day-7 blastocysts and transferred to recipients. Day-0 and Day-7 recipient plasma ($N = 36 \times 2$) and CM ($N = 36$) were analyzed by gas chromatography coupled to the quadrupole time of flight mass spectrometry (GC-qTOF). Metabolites quantified in CM and plasma were analyzed as a function to predict pregnancy at Day-40, Day-62, and birth (univariate and multivariate statistics). Subsequently, a Boolean matrix (F1 score) was constructed with metabolite pairs (one from the embryo, and one from the recipient) to combine the predictive power of embryos and recipients. Validation was performed in independent cohorts of ETs analyzed. Embryos that did not reach birth released more stearic acid, capric acid, palmitic acid, and glyceryl monostearate in CM (i.e., ($p < 0.05$, FDR < 0.05, Receiver Operator Characteristic—area under curve (ROC-AUC) > 0.669)). Within Holstein recipients, hydrocinnamic acid, alanine, and lysine predicted birth (ROC-AUC > 0.778). Asturiana de los Valles recipients that reached birth showed lower concentrations of 6-methyl-5-hepten-2-one, stearic acid, palmitic acid, and hippuric acid (ROC-AUC > 0.832). Embryonal capric acid and glyceryl-monostearate formed F1 scores generally >0.900, with metabolites found both to differ (e.g., hippuric acid, hydrocinnamic acid) or not (e.g., heptadecanoic acid, citric acid) with pregnancy in plasmas, as hypothesized. Efficient lipid metabolism in the embryo and the recipient can allow pregnancy to proceed. Changes in phenolics from plasma suggest that microbiota and liver metabolism influence the pregnancy establishment in cattle.

35.- GONZALES-BARRON, U., SANTOS-RODRIGUES, G., PIEDRA, R. B., COELHO-FERNANDES, S., OSORO, K., CELAYA, R., MAURICIO, R. S., PIRES, J., TOLSDORF, A., GESS, A., CHIESA, F., PATEIRO, M., BRUGIAPAGLIA, A., BODAS, R., BARATTA, M., LORENZO, J. M., & CADAVEZ, V. A. P. (2021). Quality attributes of lamb meat from European breeds: Effects of intrinsic properties and storage. *Small Ruminant Research, 198*(12), Art. 106354. DOI: 10.1016/j.smallrumres.2021.106354. [SPA] [Ver online]

Abstract: The sustainability of the European sheep farming sector can be ensured through the high and consistent quality of products, although the concept of meat quality, being multifaceted, is challenging to keep up by farmers and producers. The objectives of this study were (i) to evaluate quality attributes (CIE L*, a*, b*, lipid oxidation, cooking loss and Warner-Bratzler texture test) of lamb meat during cold storage originating from eight European breeds raised under their normal production systems (*n* = 292 lambs); and (ii) to elucidate the influence of storage and intrinsic properties (cold carcass weight [CCW], ultimate pH [pH_{24}], water activity [a_w], and proximate composition) on the aforementioned quality attributes measured on the 3rd, 9th and 15th day post slaughter. All meat quality properties displayed a high variability due to the breed × production systems, although, as a whole, L*, a*, b*, TBARs and cooking loss increased during storage ($P < 0.001$), whereas the tenderisation process was of non-linear decay nature ($P = 0.001$). While production system – taken across breeds – was not found to affect meat cooking losses, it heavily influenced CCW, producing intensive systems heavier ($P < 0.001$) carcasses (21.4 kg) than those raised in semi-extensive (14.9 kg) and extensive systems (13.8 kg). On the 15th day of storage, TBARs from intensive systems was significantly ($P < 0.001$) higher than those of extensively raised lambs (0.839 mg MDA/kg vs. 0.299 mg MDA/kg, respectively). Although the extensive and semi-extensive systems initially produced less tender meat ($P < 0.05$) than intensive systems, on the 15th day of storage, meat from extensive (23.4 N/cm^2) and semi-extensive systems (24.4 N/cm^2) presented as good tenderness quality ($P < 0.10$) as that of the intensive systems (22.1 N/cm^2). Within breed × production systems, higher CCW or intramuscular fat was associated to darker meat ($P < 0.001$ and $P = 0.014$), higher redness ($P < 0.001$ for both), greater lipid oxidation ($P = 0.016$ and $P < 0.001$), and lower slope ($P = 0.014$ and $P = 0.004$) and force ($P = 0.027$ and $P = 0.001$). The only intrinsic property that heavily affected most of the quality attributes was ash content, since meat of higher ash content presented higher luminosity ($P = 0.008$), and lower redness ($P < 0.001$), yellowness ($P < 0.001$), TBARS ($P < 0.001$), cooking loss ($P = 0.002$) and tenderness ($P < 0.001$). The elucidation of the impact of intrinsic properties on the lamb meat quality attributes during storage can help breeders and producers modify current flock management, feeding strategies and pre-slaughter and slaughter practices towards quality improvement or meeting particular consumers' demands.

36.- GONZÁLEZ-BLANCO, L., DIÑEIRO, Y., DÍAZ-LUIS, A., COTO-MONTES, A., OLIVÁN, M., SIERRA, V. (2021). Impact of extraction method on the detection of quality biomarkers in normal vs. DFD meat. *Foods, 10*(5), Art.1097. DOI: 10.3390/foods10051097. (OA) [SPA] [Ver online]

Abstract: The objective of this work was to demonstrate how the extraction method affects the reliability of biomarker detection and how this detection depends on the biomarker location within the cell compartment. Different extraction methods were used to study the sarcoplasmic and myofibrillar fractions of the *Longissimus thoracis et lumborum* muscle of young bulls of the Asturiana de los Valles breed in two quality grades, standard (Control) or dark, firm, and dry (DFD) meat. Protein extractability and the expression of some of the main meat quality biomarkers—oxidative status (lipoperoxidation (LPO) and catalase activity (CAT)), proteome (SDS-PAGE electrophoretic pattern), and cell stress protein (Hsp70)—were analyzed. In the sarcoplasmic fraction, buffers containing Triton X-100 showed significantly higher protein extractability, LPO, and higher intensity of high-molecular-weight protein bands, whereas the TES buffer was more sensitive to distinguishing differences in the protein pattern between the Control and DFD meat. In the myofibrillar fraction, samples extracted with the lysis buffer showed significantly higher protein extractability, whereas samples extracted with the non-denaturing buffer showed higher results for LPO, CAT, and Hsp70, and higher-intensity bands in the electrophoretic pattern. These findings

highlight the need for the careful selection of the extraction method used to analyze the different biomarkers considering their cellular location to adapt the extractive process.

37.- GOYACHE, F., FERNÁNDEZ, I., TAPSOBA, A. S. R., TRAORE, A., MENÉNDEZ-ARIAS, N. A., & ÁLVAREZ, I. (2021). Functional characterization of copy number variations regions in Djallonke sheep. *Journal of Animal Breeding and Genetics*, *138*(5), 600-612. [GRA] [Ver online]

Abstract: A total of 184 Djallonké (West African Dwarf) sheep of Burkina Faso were analysed for Copy Number Variations (CNV) using Ovine 50 K SNP BeadChip genotyping data and two different CNV calling platforms: PennCNV and QuantiSNP. Analyses allowed to identify a total of 63 candidate Copy Number Variations Regions (CNVR) on 11 different ovine chromosomes covering about 82.5 Mb of the sheep genome. Gene-annotation enrichment analysis allowed to identify a total of 751 potential candidate ovine genes located in the candidate CNVR bounds. Functional annotation allowed to identify five statistically significant Functional Clusters (FC; enrichment factor > 1.3) involving 61 candidate genes. All genes forming significantly enriched FC were located on ovine chromosome (OAR) 21. FC1 (22 genes including *PAG4* and *PAG6*) and FC5 (three genes: *CTSC*, *CTSW* and *CTSF*), coding proteases (peptidases and cathepsins, respectively), were involved in reproductive performance and modulation of gestation. Both FC3 and FC4 were involved in inflammatory and immunologic response through coding serum amyloid A and B-box-type zinc finger proteins, respectively. Finally, FC2 consisted of 27 genes (including *OR10G6* and *OR8B8*) involved in olfactory receptor activity, key for animals adapting to new food resources. CNVR identified on at least 15% of individuals were considered CNVR hotspots and further overlapped with previously reported quantitative trait loci (QTL). CNVR hotspots spanning genes putatively involved with lipid metabolism (*SKP1*, *TCF7*, *JADE2*, *UBE2B* and *SAR1B*) and differential expression in mammary gland (*SEC24A* and *CDKN2AIPNL*) on OAR5 and dairy traits (*CCDC198* and *SLC35F4*) on OAR7 overlapped with QTL associated with lipid metabolism, milk protein yield and milk fat percentage. Information obtained from local sheep populations naturally adapted to harsh environments contributes to increase our understanding of the genomic importance of CNV.

38.- GOYACHE, F., PÉREZ-PARDAL, L., FERNÁNDEZ, I., TRAORE, A., MENÉNDEZ-ARIAS, N. A., & ÁLVAREZ, I. (2021). Ancient autozygous segments subject to positive selection suggest adaptive immune responses in West African cattle. *Gene, 803*, Art.145899. DOI:10.1016/j.gene.2021.145899. [GRA] [Ver online]

Abstract: Small-sized and trypanotolerant West African taurine (*Bos taurus*) cattle are a unique case of human-mediated process of adaptation to a challenging environment. Extensive gene flow with Sahelian zebu (*B. indicus*), bigger and with some resistance to tick attack, occurred for centuries and allowed the apparition of stable crossbred populations (sanga) having intermediate characteristics. Up to 237 individuals belonging to 10 different taurine, zebu and sanga cattle populations sampled in Benin, Burkina Faso and Niger were typed using the BovineHD BeadChip of Illumina to identify signatures of selection, assessed using three different Extended-Haplotype-Homozygosity-based statistics, overlapping with ancient, originated 1024 or 2048 generations ago, Homozygosity-By-Descent segments in the cattle genome. Candidate genomic regions were defined ensuring their importance within cattle type and using zebu as reference. Functional annotation analysis identified four statistically significant

Annotation Clusters in taurine cattle (from ACt1 to ACt4), one (ACs1) in sanga, and another (ACz1) in zebu cattle, fitting well with expectations. ACt1 included genes primarily associated with innate immunity; ACt2 involved bitter taste receptor genes of importance to adaptation to changing environments; ACt3 included 68 genes coding ATP-binding proteins, some of them located on trypanotolerance-related QTL regions, that can partially underlie immune response and the additive mechanism of trypanotolerance; ACt4 was associated with growth and small size (*NPPC* gene); ACs1 included genes involved in immune response; and ACz1 is related with ectoparasite resistance. Our results provide a new set of genomic areas and candidate genes giving new insights on the genomic impact of adaptation in West African cattle.

39.- HAMBÄCK, P. A., CIRTWILL, A., GARCÍA, D., GRUDZINSKA-STERNO, M., MIÑARRO, M., TASIN, M., YANG, X., & SAMNEGÅRD, U. (2021). More intraguild prey than pest species in arachnid diets may compromise biological control in apple orchards. *Basic and Applied Ecology 57*, 1-13. DOI: 10.1016/j.baae.2021.09.006. (OA) [CHF] [Ver online]

Abstract: Understanding the full diet of natural enemies is necessary for evaluating their role as biocontrol agents, because many enemy species do not only feed on pests but also on other natural enemies. Such intraguild predation can compromise pest control if the consumed enemies are actually better for pest control than their predators. In this study, we used gut metabarcoding to quantify diets of all common arachnid species in Swedish and Spanish apple orchards. For this purpose, we designed new primers that reduce amplification of arachnid predators while retaining high amplification of all prey groups. Results suggest that most arachnids consume a large range of putative pest species on apple but also a high proportion of other natural enemies, where the latter constitute almost a third of all prey sequences. Intraguild predation also varied between regions, with a larger content of heteropteran bugs in arachnid guts from Spanish orchards, but not between orchard types. There was also a tendency for cursorial spiders to have more intraguild prey in the gut than web spiders. Two groups that may be overlooked as important biocontrol agents in apple orchards seem to be theridiid web spiders and opilionids, where the latter had several small-bodied pest species in the gut. These results thus provide important guidance for what arachnid groups should be targets of management actions, even though additional information is needed to quantify all direct and indirect interactions occurring in the complex arthropod food webs in fruit orchards.

40.- HUTCHINSON, L.A., OLIVER, T.H., BREEZE, T.D., BAILES, E.J., BRUENJES, L., CAMPBELL, A., ERHARDT, A., DE GROOT, G.A., FOLDESI, R., GARCÍA, D., GOULSON, D., HAINAUT, H., HAMBÄCK, P., HOLZSCHUH, A., JAUKER, F., KLATT, B.K., KLEIN, A-M., KLEIJN, D., KOVÁCS-HOSTYÁNSZKI, A., KRIMMER, H., MCKERCHAR, M., MIÑARRO, M., PHILIPS, B., POTTS, S.G., PUFAL, G., RADZEVIČIŪTĖ, R., ROBERTS, S.P.M., SAMNEGÅRD, U., SCHULZE, J., SHAW, R.F., TSCHARNTKE, T., VEREECKEN, N.J., WESTPHAL, C., WIETZKE, A., WOODCOCK, B.A., & GARRATT, M.P.D. (2021). Using field survey and ecological trait data to determine the bee pollinators of crops in Great Britain. *Agriculture,Ecosystems and Environment, 315*, Art. 107447. DOI: 10.1016/j.agee.2021.107447. [CHF] [Ver online]

Abstract: The importance of wild bees for crop pollination is well established, but less is known about which species contribute to service delivery to inform agricultural management, monitoring and conservation. Using sites in Great Britain as a case study, we use a novel qualitative approach combining ecological information and field survey

data to establish a national list of crop pollinating bees for four economically important crops (apple, field bean, oilseed rape and strawberry). A traits data base was used to establish potential pollinators, and combined with field data to identify both dominant crop flower visiting bee species and other species that could be important crop pollinators, but which are not presently sampled in large numbers on crops flowers. Whilst we found evidence that a small number of common, generalist species make a disproportionate contribution to flower visits, many more species were identified as potential pollinators, including rare and specialist species. Furthermore, we found evidence of substantial variation in the bee communities of different crops. Establishing a national list of crop pollinators is important for practitioners and policy makers, allowing targeted management approaches for improved ecosystem services, conservation and species monitoring. Data can be used to make recommendations about how pollinator diversity could be promoted in agricultural landscapes. Our results suggest agri-environment schemes need to support a higher diversity of species than at present, notably of solitary bees. Management would also benefit from targeting specific species to enhance crop pollination services to particular crops. Whilst our study is focused upon Great Britain, our methodology can easily be applied to other countries, crops and groups of pollinating insects.

41.- JUSTE, R. A., GEIJO, M. V., ELGUEZABAL, N., SEVILLA, I. A., ALONSO-HEARN, M., & GARRIDO, J. M. (2021). Paratuberculosis vaccination specific and non-specific effects on cattle lifespan. *Vaccine, 39*(11), 1631-1641. [SA] [Ver online]

Abstract: Records of cattle vaccination against paratuberculosis (PTB) have been analyzed to determine whether or not non-specific effect (NSE) on overall mortality similar to that observed in BCG vaccinated humans occurs in animals. The results of a previously reported slaughterhouse study on PTB prevalence were used as a reference on the age incidence of advanced patent (clinical) epidemio-pathogenic forms. In the proper vaccine study, cows in 30 cattle farms in the Basque Country, Spain were followed-up for between 1 and 13 years. Vaccinated groups were composed by 1008 (592 right-censored) animals younger than 3 months treated as calves and by 3761 (3160 right-censored) vaccinated at any older age. Controls were 339 (157 right-censored) and 4592 (2213 right-censored) age matched animals, respectively. Individual last year presence in the annual testing was considered age at culling or death. A survival analysis was carried out according age at vaccination of vaccinated versus non-vaccinated animals. PTB age incidence in the slaughterhouse study was subtracted from the difference between vaccinated and non-vaccinated animals at the same age in order to estimate PTB-specific and non-specific effects. The maximum difference was observed at the 2–3 years interval with a 33.9% mortality reduction in the calf vaccinated group. This corresponded also with the maximum NSE that was 24.5% for a PTB incidence of 9.5%. Overall, vaccination afforded to calves a 26.5% yearly mortality protection, split between 11.1% PTB-specific and 15.4% NSE. These results support a NSE on total mortality associated with PTB vaccination that appeared to persist for up to 6–7 years. This confirms for the first time in an animal field study the innate immune system memory predicted by the recently proposed trained immunity theory. Contrasting the literature, no deleterious effects of killed vaccines on females were observed. Mortality reduction would offset vaccination costs and could improve livestock systems efficiency and potentially reduce antibiotic use.

42.- LLAVANERA, M., RIBAS-MAYNOU, J., DELGADO-BERMUDEZ, A., RECUERO, S., MUINO, R., HIDALGO, C. O., TAMARGO, C., BONET, S., MATEO-OTERO, Y. & YESTE, M. (2021). Sperm chromatin condensation as an in vivo fertility biomarker in bulls: a flow cytometry approach. *Journal of Animal Science and Biotechnology,12*(1), Art. 115. DOI: 10.1186/s40104-021-00634-7. (OA) [SRA] [Ver online]

Abstract: Background : Genetic selection in cattle has been directed to increase milk production. This, coupled to the fact that the vast majority of bovine artificial inseminations (AI) are performed using cryopreserved sperm, have led to a reduction of fertility rates over the years. Thus, seeking sensitive and specific sperm biomarkers able to predict fertility rates is of vital importance to improve cattle reproductive efficiency. In humans, sperm chromatin condensation evaluated through chromomycin A3 (CMA3) has recently been purported to be a powerful biomarker for sperm functional status and male infertility. The objectives of the present study were: a) to set up a flow cytometry method for simultaneously evaluating chromatin condensation and sperm viability, and b) to test whether this parameter could be used as a predictor of *in vivo* fertility in bulls. The study included pools of three independent cryopreserved ejaculates per bull from 25 Holstein males. Reproductive outcomes of each sire were determined by non-return rates, which were used to classify bulls into two groups (highly fertile and subfertile).

Results: Chromatin condensation status of bovine sperm was evaluated through the combination of CMA3 and Yo-Pro-1 staining and flow cytometry. Sperm quality parameters (morphology, viability, total and progressive motility) were also assessed. Pearson correlation coefficients and ROC curves were calculated to assess their capacity to predict *in vivo* fertility. Sperm morphology, viability and total motility presented an area under the ROC curve (AUC) of 0.54, 0.64 and 0.68, respectively ($P > 0.05$), and thus were not able to discriminate between fertile and subfertile individuals. Alternatively, while the percentage of progressively motile sperm showed a significant predictive value, with an AUC of 0.73 ($P = 0.05$), CMA3/Yo-Pro-1 staining even depicted superior results for the prediction of *in vivo* fertility in bulls. Specifically, the percentage of viable sperm with poor chromatin condensation showed better accuracy and precision to predict *in vivo* fertility, with an AUC of 0.78 ($P = 0.02$).

Conclusions: Chromatin condensation evaluated through CMA3/Yo-Pro-1 and flow cytometry is defined here as a more powerful tool than conventional sperm parameters to predict bull *in vivo* fertility, with a potential ability to maximising the efficiency of dairy breeding industry.

43.- MARTÍNEZ-RODERO, I., GARCÍA-MARTÍNEZ, T., ORDOÑEZ-LEÓN, E. A., VENDRELL-FLOTATS, M., HIDALGO, C. O., ESMORIS, J., MENDIBIL, X., AZCÁRATE, S., LÓPEZ-BÉJAR, M., YESTE, M., & MOGAS, T. (2021). A shorter equilibration period improves post-warming outcomes after vitrification and in straw dilution of in vitro-produced bovine embryos. *Biology-Basel, 10*(2), Art.142. (OA) [SRA] [Ver online]

Abstract: This study was designed to the optimize vitrification and in-straw warming protocol of in vitro-produced bovine embryos by comparing two different equilibration periods, short equilibrium (SE: 3 min) and long equilibrium (LE: 12 min). Outcomes recorded in vitrified day seven (D7) and day eight (D8) expanded blastocysts were survival and hatching rates, cell counts, apoptosis rate, and gene expression. While survival rates at 3 and 24 h post-warming were reduced ($p < 0.05$) after vitrification, the hatching rates of D7 embryos vitrified after SE were similar to the rates recorded in fresh non-vitrified blastocysts. The hatching rates of vitrified D8 blastocysts were lower ($p < 0.05$) than of fresh controls regardless of treatment. Total cell count, and inner cell mass and trophectoderm cell counts were similar in hatched D7 blastocysts vitrified after SE and fresh blastocysts, while vitrified D8 blastocysts yielded lower values regardless of treatment. The apoptosis rate was significantly higher in both treatment groups compared to fresh controls, although rates were lower for SE than LE. No differences emerged in *BAX*, *AQP3*, *CX43,* and *IFNτ* gene expression between the treatments, whereas a significantly greater abundance of *BCL2L1* and *SOD1* transcripts

was observed in blastocysts vitrified after SE. A shorter equilibration vitrification protocol was found to improve post-warming outcomes and time efficiency after in-straw warming/dilution.

44.- MARTÍNEZ-SASTRE, R., PEÑA, R., GONZÁLEZ-IBÁÑEZ, A., GARCÍA, D., & MIÑARRO, M. (2021). Top-down and bottom-up regulation of codling moth populations in cider apple orchards. *Crop Protection, 143.* Art. 105545. [CHF] [Ver online]

Abstract: The success of biological control by natural enemies in agricultural crops relies on an understanding of the trophic interactions between natural enemies, pests and host plants. Top-down and bottom-up trophic effects, together with potential landscape and local-scale factors, may regulate pest populations. For two years, we analyzed codling moth populations (*Cydia pomonella*), their crop damage and their parasitoid communities in 26 low-input cider apple orchards in northern Spain. Codling moth abundance was estimated from overwintering larvae sampled with cardboard traps on trees, parasitism was estimated from parasitoids emerged from lab-reared moth larvae, and pest damage was assessed in apples before ripening. Codling moth abundance differed between orchards across years, and was positively correlated with apple production and the cover of apple plantations in the surrounding landscape. The effects of the apple production on codling moth abundance suggest bottom-up pest regulation. Apple damage in individual orchards reached 71%, but decreased with apple production, indicating codling moth satiation. Seven parasitoid species were recorded on codling moth larvae. Parasitism rate in individual orchards reached 42.5% of codling moth larvae. The number of parasitized larvae per orchard was positively related to parasitoid richness, but also to codling moth abundance, suggesting simultaneous top-down and bottom-up effects between parasitoids and pest. This study highlights the need to tackle the whole parasitoid-pest-plant system in order to better manage codling moth damage in orchards. The conservation of complementary parasitoid species through biodiversity-friendly actions should be combined with the control of apple production at the orchard- and landscape scale.

45.- MIÑARRO, M., & GARCÍA, D. (2021). Complementary contribution of wild bumblebees and managed honeybee to the pollination niche of an introduced blueberry crop. *Insects 12*(7), Art. 595. DOI: 10.3390/insects12070595. (OA) [CHF] [Ver online]

Abstract: The entomophilous pollination niche (abundance, phenotypic traits, foraging behaviours and environmental tolerances of insect pollinators) helps to understand and better manage crop pollination. We apply this niche approach to assess how an entomophilous crop (blueberry, *Vaccinium ashei*) can be expanded into new territories (i.e., northern Spain) far from their original area of domestication (North America). Insect visits to blueberry flowers were monitored in a plantation on 12 different days, at 8 different times during day and covering various weather conditions. Abundance, visitation rate, pollen gathering behaviour, and frequency of inter-plant and inter-row movements were recorded. The pollinator assemblage was basically composed of one managed honeybee species (50.8% of visits) and three native bumblebee species (48.3%). There was a marked pattern of seasonal segregation throughout bloom, with bumblebees dominating the early bloom and honeybee the late bloom. Pollinators also segregated along gradients of daily temperature and relative humidity. Finally, the two pollinator types differed in foraging behaviour, with bumblebees having a visitation rate double that of honeybee, collecting pollen more frequently and changing plant and row more frequently. The spatio-temporal and functional complementarity between honeybee and bumblebees suggested here encourages the consideration of an integrated crop pollination strategy for blueberries, based on the concurrence of both wild and managed bees.

46.- MORÁN, F., MARCO-NOALES, E., LANDERAS, E., ROSELLO, M., ABELLEIRA, A., GONZÁLEZ, A. J., & LÓPEZ, M. M. (2021). Polyphasic analysis of isolates from kiwifruit reveal new genetic lineages of Pseudomonas syringae pv. actinidifoliorum look-alike. *Agronomy-Basel, 11*(12), Art. 2464. DOI: 10.3390/agronomy11122464. (OA) [CHF] [Ver online]

Abstract: Currently, kiwifruit cultivation arouses great economic interest in the agricultural sector in several countries of the European Union due to high consumer demand and good results achieved in terms of production potential and fruit quality. One of the main bacterial species that cause yield losses in kiwifruit plants is *Pseudomonas syringae*. Diseases such as bacterial canker, caused by pathovar (pv.) *actinidiae*; floral bud necrosis caused by pv. *syringae* and leaf spots caused by pv. *actinidifoliorum* (Pfm) are clear examples. Between 2014 and 2017, in the main kiwifruit producing areas in the north and east of Spain, several surveys were carried out in search of these pathogens. Analyses realized from symptomatic and asymptomatic plants of *Actinidiadeliciosa* revealed the existence of new bacterial isolates close to Pfm. These new isolates werelow virulence pathogens similar to Pfm but belonging to a new group of *P. syringae* that affected the leaves of *A. chinensis* var. *deliciosa.* This study focused on the characterization and classification of these new isolates by a polyphasic approach in order to provide more information for understanding how the different populations of *P.syringae* affecting kiwifruit. They had the phenotypic characteristics of Pfm but by molecular approaches, they constituted a supported genetic lineage closely-related to Pfm independent of the five lineages described so far. This work revealed the great diversity found in *P. syringae* species affecting kiwifruit plants and supports the hypothesis that Pfm is a low virulence pathogen which is long established in Europe.

47.- OJEDA-MARIN, C., CERVANTES, I., MORENO, E., GOYACHE, F., & GUTIERREZ, J. P. (2021). Breeding strategies to optimize effective population size in low census captive populations: The case of *Gazella cuvieri*. *Animals, 11*(6), Art. 1559. DOI: 10.3390/ani11061559. (OA) [GRA] [Ver online]

Abstract: Small-sized animal populations can undergo significant loss of genetic variability that can lead to their extinction. Therefore, studies on animal breeding have focused on mating systems for minimizing the disappearance of genetic variability. The main objective of this study was to compare, using computer simulations, the performance of different breeding schemes to limit the loss of genetic diversity in small-sized populations. This objective was achieved by monitoring the evolution of the effective population size obtained by 23 strategies throughout 20 generations in two populations of *Gazella cuvieri*. The scenarios were designed with different assumptions, in both reference subpopulations, regarding: the use of parents coancestry or offspring coancestry, the use of their increases or the coefficients themselves, and the number of males and females involved. Computations were performed using an experimental module of Endog v4.9 developed for this purpose. The results of the study showed that strategies for minimizing the coancestry of the parents were better in the short term; however, these strategies were worse in the long term. Minimizing the average coancestry of the offspring was a better approach in the long term. Nevertheless, in both populations, the best results were obtained when both the coancestry of the

parents and the coancestry of the offspring were weighted at 5% each and neither males nor females were assumed to contribute to the next generation. In any case, not all strategies had the same evolutionary pattern throughout generations in both populations. The current results show that neither traditional nor new strategies have any general use. Therefore, it is important to carefully test these strategies before applying them to different populations with different breeding needs under different conditions, such as different generation intervals, and different natural breeding systems such as monogamy or polygyny.

48.- OLEAGA, A., BALSEIRO, A., ESPÍ, A., & ROYO, L. J. (2021). Wolf (*Canis lupus*) as canine adenovirus type 1 (CAdV-1) sentinel for the endangered cantabrian brown bear (*Ursus arctos arctos*). *Transboundary and Emerging Diseases.* DOI: 10.1111/tbed.14010. [SA] [NPF] [Ver online]

Abstract: Canine adenovirus type 1 (CAdV-1) causes infectious canine hepatitis (ICH) and has recently been described as a cause of death among endangered populations of European brown bear (*Ursus arctos arctos*) in the Cantabrian mountain range in Asturias, Spain. Sympatric wild and domestic carnivores can act as reservoirs of the virus and likely spread it into the environment and subsequently transmit it to brown bears. The present work investigates the prevalence and geo-temporal distribution of CAdV-1 among free-ranging wolves (*Canis lupus*) in Asturias from 2009 to 2018, during which three fatal cases of ICH were reported among brown bears in the region. A total of 149 wolves were analysed in this study, of which 21 (14%) were found to have CAdV-1 DNA based on real-time polymerase chain reaction (RT-PCR) of spleen samples. Prevalence of the virus was similar between males and females. All but one of the 20 CAdV-1-positive animals of estimable age were younger than 2 years, and only one of the 46 adult animals (>2 years) tested positive. Prevalence was highest in the western area of Asturias and during 2010 and 2011. Our results confirm that CAdV-1 is circulating in Asturian free-ranging wolves, supporting their possible role as virus reservoirs and sentinels in the region of this emerging disease in brown bears.

49.- OLEAGA, Á., VÁZQUEZ, C. B., ROYO, L. J., BARRAL, T. D., BONNAIRE, D., ARMENTEROS, J. Á., RABANAL, B., GORTÁZAR, C., & BALSEIRO, A. (2021). Canine distemper virus in wildlife in southwestern Europe. *Transboundary and Emerging Diseases*. DOI: 10.1111/tbed.14323. (OA) [SA] [NPF] [Ver online]

Abstract: Multi-host pathogens emerging and re-emerging at the wildlife–domestic animal interface affect wildlife management and conservation. This is the case of canine distemper virus (CDV), a paramyxovirus closely related to human measles virus and rinderpest virus of cattle. With an area of 10,603 km^2, Asturias region in Atlantic Spain is a hotspot of carnivore diversity, which includes the largest Eurasian brown bear (*Ursus arctos arctos*) population and one of the largest wolf (*Canis lupus*) populations in south-western Europe. In 2020–2021, we recorded mortality due to distemper in four carnivore species including three mustelids (Eurasian badger *Meles meles*, European marten *Martes martes* and European polecat *Mustela putorius*) and one canid (red fox, *Vulpes vulpes*). Clinical signs and pathology were similar across species and consistent with the emergence of a highly pathogenic viral strain, with CDV antigen mainly located in the central nervous system, lungs, spleen and lymph nodes. A molecular study in eight wild carnivore species, also including the Iberian wolf, Eurasian brown bear, American mink (*Neovison vison*) and stone marten (*Martes foina*), revealed 19.51% (16/82) of positivity. Phylogenetic analysis demonstrated that CDV belonged to the previously described European lineage. A retrospective serosurvey (2008–2020) showed a high seroprevalence of CDV antibodies (43.4%) in 684 analyzed badgers, indicating a long-term though not stable viral circulation in this multi-host community. The possible triggers of the 2020–2021 outbreak and the implications for carnivore management and conservation are discussed.

50.- OSTERMAN, J., LANDAVERDE-GONZÁLEZ, P., GARRATT, M.P.D., GEE, M., MANDELIK, Y., LANGOWSKA, A., MIÑARRO, M. COLE, L.J., EERAERTS, M., BEVK, D., AVRECH, O., KOLTOWSKI, Z., TRUJILLO-ELISEA, F.I., PAXTON, R.J., BOREUX, V., SEYMOUR, C.L., & . HOWLETT, B.G. (2021). On-farm experiences shape farmer knowledge, perceptions of pollinators, and management practices. *Global Ecology and Conservation*, *32,* Art. e019492021. DOI: 10.1016/j.gecco. (OA) [CHF] [Ver online]

Abstract: Mitigating pollinator declines in agriculturally dominated landscapes to safeguard pollination services requires the involvement of farmers and their willingness to adopt pollinator-friendly management. However, farmer knowledge, perceptions, and actions to support on-farm pollinators and their alignment with science-based knowledge and recommendations are rarely evaluated. To close this knowledge gap, we interviewed 560 farmers from 11 countries around the world, cultivating at least one of four widely grown pollinator-dependent crops (apple, avocado, kiwifruit, oilseed rape). We particularly focused on non-bee crop pollinators which, despite being important pollinators of many crops, received less research attention than bees. We found that farmers perceived bees to be more important pollinators than other flower-visiting insects. However, around 75% of the farmers acknowledged that non-bees contributed to the pollination of their crops, seeing them as additional pollinators rather than substitutes for bees. Despite farmers rating their own observations as being most important in how they perceived the contribution of different crop pollinator taxa, their perception aligned closely with results from available scientific studies across crops and countries. Farmer perceptions were also linked with their pollinator management practices, e.g. farmers who used managed bees for crop pollination services (more than half the farmers) rated these managed bees as particularly important. Interestingly, their willingness to establish wildflower strips or manage hedgerows to enhance pollinator visitation was linked to their ecological knowledge of non-bees or to government subsidies. Farmers adapted practices to enhance pollination services depending on the crop, which indicates an understanding of differences in the pollination ecology of crops. Almost half of the farmers had changed on-farm pollination management in the past 10 years and farm practices differed greatly between countries. This suggests integrated crop pollination measures are being adapted by farmers to reach best pollinator management practices. Our findings highlight the importance of studying local knowledge as a key to co-design locally-adapted measures to facilitate pollinator-integrated food production as ecological intensification tools.

51.- PANDO BEDRIÑANA, R., PICINELLI LOBO, A., RODRÍGUEZ MADRERA, R., & SUÁREZ VALLES, B. (2021). New ciders made by an exhaustion method: an option to val-orise subproducts from the making of ice ciders. *Beverages, 7*(4), Art. 75. DOI: 10.3390/beverages7040075. (OA) [TA] [Ver online]

Abstract: Cryo-extraction (pressing of frozen apples), is one of the two freeze-enrichment systems allowed for the making of ice juices. Its ciders are often described as more complex and aromatic, however, the production yield is quite low. The Exhaustion method associated with the previous one proposes the valorisation of the

discarded apple juice fractions for the making of new ciders. Three types of apple juices and three species of yeasts (*S. bayanus*, C6; *S. cerevisiae*, Levuline-CHP, and *T. delbrueckii*, Biodiva-TD291) have been used to evaluate the Exhaustion method. The ciders obtained were analysed for chemical and volatile composition as well as sensory characteristics. The yield (%) of the Exhaustion process ranged between 24 and 37%. The yeasts promoted the fermentation at different rates, providing ciders with alcoholic degrees between 9 and 12 (%*v*/*v*), and low volatile acidities. The yeast strain significantly influenced most of the parameters analysed, whereas the raw apple juice influenced the perception of the attributes fruity, apple and butter. Although the ciders produced by Exhaustion presented significantly lower concentrations of all the volatile compounds analysed than the corresponding ice ciders obtained by Cryo-extraction, the *S. bayanus* C6 and *T. delbrueckii* TD291 gave highly valued ciders from the sensory point of view.

52.- PEÑA-FERNÁNDEZ, N., CANO-TERRIZA, D., GARCÍA-BOCANEGRA, I., HORCAJO, P., VÁZQUEZ-ARBAIZAR, P., CLEOFE-RESTA, D., PÉREZ ARROYO, B., ORTEGA MORA, L.M.,& COLLANTES-FERNÁNDEZ, E. (2021). Prevalence of bovine genital *Campylobacteriosis*, associated risk factors and spatial distribution in Spanish beef cattle based on veterinary laboratory database records. *Frontiers in Veterinary Science, 8*, Art. 750183. DOI: 10.3389/fvets.2021.750183. (OA) [SA] [Ver online]

Abstract: Bovine genital campylobacteriosis (BGC) is a sexually transmitted disease that causes early reproductive failure in natural breeding cattle that are managed extensively. The aim of this study was to assess the BGC prevalence in Spain from 2011 to 2019 using data collected cross-sectionally from the diagnostic reports issued by the SALUVET veterinary diagnostic laboratory from a total of 5,182 breeding bulls from 1,950 herds managed under “dehesa” systems (large herds within fenced pastures and all-year breeding season) or mountain systems (smaller herds with seasonal breeding management and grazing in communal mountain pastures). Infection was detected by PCR in 7.7 and 12.2% of the bulls and herds tested, respectively. The “dehesa” herd management system (OR = 2.078, $P = < 0.001$, 95% CI = 1.55–1.77), bovine trichomonosis status of the herd (OR = 1.606, $P = 0.004$, 95% CI = 1.15–2.22), and bulls ≥3 years old (OR = 1.392, $P = 0.04$, 95% CI = 1.01–1.92) were identified as risk factors associated with *Campylobacter fetus venerealis* infection. We also studied the high-risk areas for circulation of the infection in extensive beef cattle herds in Spain, showing four significant clusters in “dehesa” areas in the south-western provinces of the country and a fifth cluster located in a mountain area in northern Spain. The results obtained in the present study indicate that BGC is endemic and widely distributed in Spanish beef herds. Specifically, “dehesa” herds are at greater risk for introduction of *Cfv* based on relatively high local prevalence of the infection and the use of specific management practices**.**

53.- RIBAS-MAYNOU, J., GARCÍA-BONAVILA, E., HIDALGO, C. O., CATALÁN, J., MIRO, J., & YESTE, M. (2021). Species-Specific differences in sperm chromatin decondensation between Eutherian mammals underlie distinct lysis requirements. *Frontiers in Cell and Developmental Biology, 9*(11), Art. 669182. DOI: 10.3389/fcell.2021.669182. (OA) [SRA] [Ver online]

Abstract: Sperm present a highly particular DNA condensation that is acquired during their differentiation. Protamines are key elements for DNA condensation. However, whereas the presence of protamine 1 (P1) is conserved across mammalian species,

that of protamine 2 (P2) has evolved differentially, existing only few species that use both protamines for sperm DNA condensation. In addition, altered P1/P2 ratios and alterations in the expression of P1 have previously been associated to infertility and DNA damage disorders. On the other hand, different methods evaluating DNA integrity, such as Sperm Chromatin Dispersion (SCD) and Comet tests, need a previous complete DNA decondensation to properly assess DNA breaks. Related with this, the present study aims to analyze the resilience of sperm DNA to decodensation in different eutherian mammals. Sperm samples from humans, horses, cattle, pigs and donkeys were used. Samples were embedded in low melting point agarose and treated with lysis solutions to induce DNA decondensation and formation of sperm haloes. The treatment consisted of three steps: (1) incubation in SDS + DTT for 30 min; (2) incubation in DTT + NaCl for 30 min; and (3) incubation in DTT + NaCl with or without proteinase K for a variable time of 0, 30, or 180 min. How incubation with the third lysis solution (with or without proteinase K) for 0, 30, and 180 min affected DNA decondensation was tested through analyzing core and halo diameters in 50 sperm per sample. Halo/core length ratio was used as an indicator of complete chromatin decondensation. While incubation time with the third lysis solution had no impact on halo/core length ratios in species having P1 and P2 (human, equine and donkey), DNA decondensation of pig and cattle sperm, which only present P1, significantly ($P < 0.05$) increased following incubation with the third lysis solution for 180 min. In addition, the inclusion of proteinase K was found to accelerate DNA decondensation. In conclusion, longer incubations in lysis solution including proteinase K lead to higher DNA decondensation in porcine and bovine sperm. This suggests that tests intended to analyze DNA damage, such as halo or Comet assays, require complete chromatin deprotamination to achieve high sensitivity in the detection of DNA breaks.

54.- RODRÍGUEZ MADRERA, R., CAMPA NEGRILLO, A., SUÁREZ VALLES, B, FERREIRA FERNÁNDEZ, J.J. (2021). Phenolic content and antioxidant activity in seeds of common bean (*Phaseolus vulgaris L*.). *Foods, 10*(4), Art. 864. DOI: 10.3390/foods10040864. (OA) [TA] [CHF] [Ver online]

Abstract: Dry bean (*Phaseolus vulgaris* L.) is one of the most important pulses consumed in the world. Total phenolic content, total flavonoid content, total monomeric anthocyanin content and antioxidant capacity were determined, using ferric reducing antioxidant power and free radical scavenging activity, in 255 lines grown under the same environmental conditions. For all parameters analysed, there was a wide range of variability, with differences always above one order of magnitude. Phenolic compounds in beans with coloured coats were found to be more efficient antioxidants than those with completely white coats, and samples with more strongly coloured coats (red, cream, black, pink and brown) showed the highest antioxidant capacities. Based on the strong correlation detected between the variables, total phenolic content can be considered an appropriate indicator of antioxidant activity. The results provide a robust database for selecting those lines of greater functional and nutritional interest in terms of cultivation for direct consumption, for inclusions in food formulations or for use in future breeding programs.

55.- RODRÍGUEZ MADRERA, R., PANDO BEDRIÑANA, R., & SÚAREZ VALLES, B. (2021). Evaluation of indigenous non-*Saccharomyces* cider yeasts for use in brewing. *European Food Research and Technology, 247*, 819-828. DOI: 10.1007/s00217-020-03665-y. [TA] [Ver online]

Abstract: The non-*Saccharomyces* yeasts represent a great richness for the production of new and different types of beers. In this study, the role of eight yeast strains (*Saccharomycodes ludwigii*, *Metchnikowia pulcherrima*, *Hanseniaspora uvarum*, *Hanseniaspora osmophila* and *Brettanomyces bruxellensis*) isolated from Asturian cider and belonging to five unconventional species was evaluated. None of the strains could ferment the main wort sugars (maltose and maltotriose). Beers produced by *S. ludwigii* strains showed the highest ethanol content (21.6 g/L) and interestingly high levels of fusel alcohols, fatty esters and isoamyl acetate. Strains of *B. bruxellensis* were the only ones that produced ethyl phenols and they also generated the highest concentration of fatty acids and their ethyl esters. Sucrose was not utilised by *Metschnikowia* and *Hanseniaspora* strains, resulting in low ethanol formation (4.7 g/L), although these yeasts differed in terms of their synthesis of volatile compounds. Larger amounts of fatty acid esters were detected in beers fermented by *H. osmophyla* 10, while *H. uvarum* 62 produced more 2-phenylethyl acetate and ethyl acetate. *M. pulcherrima* 301 stands out for its low fermentative capacity and production of volatile compounds. These results show the interest of non-*Saccharomyces* strains in the production of beers with low alcohol content and their potential use in developing new beer styles in sequential or mixed inoculations with *S. cerevisiae* strains.

56.- RODRÍGUEZ MADRERA, R., & SUÁREZ VALLES, B. (2021). Analysis of cyanogenic compounds derived from mandelonitrile by ultrasound-assisted extraction and high-performance liquid chromatography in *Rosaceae* and *Sambucus* families. *Molecules, 26*, Art. 7563. DOI: 10.3390/molecules26247563. (OA) [TA] [Ver online]

Abstract: An analytical method for extraction and quantitative determination of amygdalin, prunasin, and sambunigrin in plant material is described. The method is based on extraction with high-power ultrasound (UAE), with acidified water as solvent and quantification by HPLC–DAD. The best extraction conditions were: 80% sonication amplitude, 55 s extraction time, 70% duty cycle, 0.1 g sample mass, and 10 mL acidified water (0.1% perchloric acid). Once developed, the method was validated in terms of accuracy and precision. Good linearity was obtained, with correlation coefficients exceeding 0.999 and the quantification limits ranged from 2.2 µg/g (amygdalin) to 9.6 µg/g (sambunigrin). The accuracy (recovery study) ranged between 90 and 104% and the reproducibility of the method was always <2.3% (RSD). Special attention should be paid to the ratio sample/solvent in samples with potential β-glucosidase activity to avoid degradation of the cyanogenic glycosides (CNGs). The proposed method was used to evaluate the content of CNGs in kernels of *Prunus* genera, apple seeds, apple pomace, and different plant materials of *Sambucus nigra*.

57.- ROQUER-BENI, L., ALINS, G., ARNAN, X., BOREUX, V., GARCÍA, D., HAMBÄCK, P.A., HAPPE , A-K., KLEIN, A-M., MIÑARRO , M., MODY, K., PORCEL, M., RODRIGO, A., SAMNEGÅRD, U., TASIN, M., & BOSCH, J. (2021). Management-dependent effects of pollinator functional diversity on apple pollination services: a response-effect trait approach. *Journal of Applied Ecology 58*(12), 2843-2853. DOI: 10.1111/1365-2664.14022. (OA) [CHF] [Ver online]

Abstract: Functional traits mediate the response of communities to disturbances (response traits) and their contribution to ecosystem functions (effect traits). To predict how anthropogenic disturbances influence ecosystem services requires a dual approach including both trait concepts. Here, we used a response–effect trait

conceptual framework to understand how local and landscape features affect pollinator functional diversity and pollination services in apple orchards.

We worked in 110 apple orchards across four European regions. Orchards differed in management practices. Low-intensity (LI) orchards were certified organic or followed close-to-organic practices. High-intensity (HI) orchards followed integrated pest management practices. Within each management type, orchards encompassed a range of local (flower diversity, agri-environmental structures) and landscape features (orchard and pollinator-friendly habitat cover). We measured pollinator visitation rates and calculated trait composition metrics based on 10 pollinator traits. We used initial fruit set as a measure of pollination service.

Some pollinator traits (body size and hairiness) were negatively related to orchard cover and positively affected by pollinator-friendly habitat cover. Bee functional diversity was lower in HI orchards and decreased with increased landscape orchard cover. Pollination service was not associated with any particular trait but increased with pollinator trait diversity in LI orchards. As a result, LI orchards with high pollinator trait diversity reached levels of pollination service similar to those of HI orchards.

Synthesis and applications. Pollinator functional diversity enables pollinator communities to respond to agricultural intensification and to increase pollination function. Our results show that efforts to promote biodiversity provide greater returns in low-intensity than in high-intensity orchards. The fact that low-intensity orchards with high pollinator functional diversity reach levels of pollination services similar to those of high-intensity orchards provides a compelling argument for the conversion of high-intensity into low-intensity farms.

58.- ROSA GARCIA, R., PERIC, T., CADAVEZ, V., GEß, A. D., LIMA CERQUEIRA, J. O., GONZALES-BARRON, U., & BARATTA, M. (2021). Arthropod biodiversity associated to European sheep production systems. *Small Ruminant Research, 205,* Art. 106536. DOI: 10.1016/j.smallrumres.2021.106536. [NPF] [Ver online]

Abstract: The rural territories linked to European sheep systems still cover wide areas and provide multiple ecosystems services although the current situation of the associated biodiversity is not fully understood. In this study the foliage arthropods (including pollinators), the vegetation cover and height, the number of flowers and plant species richness were evaluated in 9 sheep grazed lands from 5 EU countries with different livestock management strategies and dominant vegetation. The total abundance of arthropods, the abundance of Diptera and Heteroptera, sward height and plant species richness were higher in more extensive than in more intensively managed farms. The total abundance and the abundance of most of the orders were highest in mountain areas (MP) and lowest in improved pastures (IMP) whereas the total arthropod richness showed no differences and the richness of pollinators was lower in IMP than in MP ($p < 0.01$) and semi-natural pastures (SN, $p < 0.01$). The grass cover was higher in IMP than in the rest of the areas whereas forb cover was higher in SN than in IMP ($p < 0.01$). The plant species richness peaked in MP whereas the number of flowers showed no significant differences. Sward height correlated positively with forb cover, plant species richness, the richness of the whole arthropod community, the abundance of several orders like Araneae, Diptera or Homoptera, as well as with the richness of the pollinator community. The community composition of the total arthropod fauna ($p < 0.01$) and the pollinators in particular ($p < 0.05$) differed between management strategies and more diverse groups were linked to the areas under more extensive management. Both communities (total and pollinators) also differed in composition between the types of vegetation ($p < 0.01$) and less diverse assemblages with low abundant taxa were associated to IMP and SN whereas more diverse groups were linked to MP and grassland-forest (WP) in both cases. A better understanding of the flora-fauna dynamics in sheep grazed pasturelands is essential for the proper conservation of the biodiversity and other ecosystem services, as well as for the maintenance of sustainable sheep systems relying on the natural resources.

59.- SALMAN, A., CAAMAÑO, J. N., FERNÁNDEZ-ALEGRE, E., HIDALGO, C. O., NADRI, T., TAMARGO, C., FUEYO, C., FERNÁNDEZ, A., MERINO, M. J., & MARTÍNEZ-PASTOR, F. (2021). Supplementation of the BIOXcell extender with the antioxidants crocin, curcumin and GSH for freezing bull semen. *Research in Veterinary Science, 136*, 444-452. DOI: 10.1016/j.rvsc.2021.03.025. [SRA] [Ver online]

Abstract: Semen cryopreservation is routine in cattle, but the results of artificial insemination need improvement. A strategy to these aims is the supplementation of the freezing extender with novel antioxidants. This study aimed at testing the natural antioxidants curcumin and crocin as supplements to the commercial extender BIOXcell for freezing semen from 8 Holstein bulls. We tested curcumin at 0.05 and 0.1 mM (CU0.05, CU0.1) and crocin at 0.5 and 1.5 mM (CR0.5, CR1.5), with 0.5 mM reduced glutathione (GSH0.5) as reference, and a control (CTL, without supplementation). The samples were evaluated post-thawing and after 5 h at 38 °C by CASA for motility and flow cytometry for viability, apoptotic, capacitation, acrosomal status, cytoplasmic and mitochondrial reactive oxygen species (ROS) production, and chromatin status (SCSA). Control and GSH0.5 showed similar results, possibly because of the good protection from BIOXcell. CU0.05 and CU0.1 showed little effects but increased cytoplasmic ROS production and motility ALH. CR0.5 and CR1.5 decreased viability and increased apoptotic features significantly post-thawing and after the incubation, resulting in lower motility (significant after the incubation) but decreasing SCSA %HDS (loose chromatin). Whereas crocin at these concentrations seems incompatible with BIOXcell, maybe because of a prooxidant activity, curcumin use merits further research, considering the elevation of ROS with no significant negative effects.

60.- SENTANDREU, E., FUENTE-GARCÍA, C., PARDO, O., OLIVÁN, M., LEÓN, N., ALDAI, N., YUSÁ, V., & SENTANDREU, M. A. (2021). Protein biomarkers of bovine defective meats at a glance: Gel-free hybrid quadrupole-Orbitrap analysis for rapid screening. *Journal of Agricultural and Food Chemistry, 69*(26), 7478-7487. DOI: 10.1021/acs.jafc.1c02016. (OA) [SPA] [Ver online]

Abstract: An understanding of biological mechanisms that could be involved in the stress response of animal cattle prior to slaughter is critical to create effective strategies aiming at the production of high-quality meat. The sarcoplasmic proteome of directly extracted samples from normal and high ultimate pH (pHu) meat groups was studied through a straightforward gel-free strategy supported by liquid chromatography hybrid quadrupole-Orbitrap high-resolution mass spectrometry (LC-HRMS) analysis. A stepped proteomic pipeline combining rapid biomarker hunting supported by qualitative protein Mascot scores followed by targeted label-free peptide quantification revealed 26 descriptors that characterized meat groups assayed. The functional study of the proposed biomarkers suggested their relevant role in metabolic, chaperone/stress-related, muscle contractility/fiber organization, and transport activities. The efficiency, flexibility, rapidity, and easiness of the methodology proposed can positively contribute to the creation of innovative proteomic alternatives addressing meat quality assessment.

61.- SIERRA, V., GONZÁLEZ-BLANCO, V., DIÑEIRO, Y., DÍAZ, F., GARCÍA-ESPINA, M.J., COTO-MONTES, A., GAGAOUA, M., & OLIVÁN. M. (2021). New insights on the impact of cattle handling on post-mortem myofibrillar muscle proteome and meat tenderization. *Foods,10*(12), Art. 3115. DOI: 10.3390/foods10123115. (OA) [SPA] [Ver online]

Abstract: This study investigated the effect of different cattle management strategies at farm (Intensive vs. Extensive) and during transport and lairage (mixing vs. non-mixing with unfamiliar animals) on the myofibrillar subproteome of *Longissimus thoracis* et *lumborum* (LTL) muscle of “Asturiana de los Valles” yearling bulls. It further aimed to study the relationships with beef quality traits including pH, color, and tenderness evaluated by Warner–Bratzler shear force (WBSF). Thus, comparative proteomics of the myofibrillar fraction along meat maturation (from 2 h to 14 days *post-mortem*) and different quality traits were analyzed. A total of 23 protein fragments corresponding to 21 unique proteins showed significant differences among the treatments ($p < 0.05$) due to any of the factors considered (Farm, Transport and Lairage, and *post-mortem* time ageing). The proteins belong to several biological pathways including three structural proteins (MYBPC2, TNNT3, and MYL1) and one metabolic enzyme (ALDOA) that were affected by both Farm and Transport/Lairage factors. ACTA1, LDB3, and FHL2 were affected by Farm factors, while TNNI2 and MYLPF (structural proteins), PKM (metabolic enzyme), and HSPB1 (small Heat shock protein) were affected by Transport/Lairage factors. Several correlations were found between the changing proteins (PKM, ALDOA, TNNI2, TNNT3, ACTA1, MYL1, and CRYAB) and color and tenderness beef quality traits, indicating their importance in the determination of meat quality and their possible use as putative biomarkers.

62.- URAL, S., MENÉNDEZ-MIRANDA M., SALZANO, G., MATHURIN J., NESLIHAN AYBEKE, E., DENISET-BESSEAU, A, DAZZI, A., PORCINO, M., MARTINEAU-CORCOS, C., & GREF, R., (2021). Compartmentalized polymeric nanoparticles deliver Vancomycin in a pH-Responsive manner. *Pharmaceutics,13*(12), Art.1992. DOI: 10.3390/pharmaceutics13121992. (OA) [NPF] [Ver online]

Abstract: Vancomycin (VCM) is a last resort antibiotic in the treatment of severe Gram-positive infections. However, its administration is limited by several drawbacks such as: strong pH-dependent charge, tendency to aggregate, low bioavailability, and poor cellular uptake. These drawbacks were circumvented by engineering pH-responsive nanoparticles (NPs) capable to incorporate high VCM payload and deliver it specifically at slightly acidic pH corresponding to infection sites. Taking advantage of peculiar physicochemical properties of VCM, here we show how to incorporate VCM efficiently in biodegradable NPs made of poly(lactic-co-glycolic acid) and polylactic acid (co)polymers. The NPs were prepared by a simple and reproducible method, establishing strong electrostatic interactions between VCM and the (co)polymers’ end groups. VCM payloads reached up to 25 wt%. The drug loading mechanism was investigated by solid state nuclear magnetic resonance spectroscopy. The engineered NPs were characterized by a set of advanced physicochemical methods, which allowed examining their morphology, internal structures, and chemical composition on an individual NP basis. The compartmentalized structure of NPs was evidenced by cryogenic transmission electronic microscopy, whereas the chemical composition of the NPs’ top layers and core was obtained by electron microscopies associated with energy-dispersive X-ray spectroscopy. Noteworthy, atomic force microscopy coupled to infrared spectroscopy allowed mapping the drug location and gave semiquantitative information about the loadings of individual NPs. In addition, the NPs were stable upon storage and did not release the incorporated drug at neutral pH. Interestingly, a slight acidification of the medium induced a rapid VCM release. The compartmentalized NPs could find potential applications for controlled VCM release at an infected site with local acidic pH.

63.- VICENTE, F., ELOUADAF, D., SÁNCHEZ-VERA, A., SOLDADO, A., DE LA TORRE-SANTOS, S., & MARTÍNEZ-FERNÁNDEZ, A. (2021). The dairy cow slurry composition used as organic fertilizer is influenced by the level and origin of the dietary protein. *Animals, 11*(10), Art. 2812. DOI: 10.3390/ani11102812. (OA) [NPF] [Ver online]

Abstract: Less than 30% of dairy cattle's nitrogen ingested is retained in milk. Therefore, large amounts of nitrogen can be excreted in manure and urine with a potential environmental impact. In addition, some legume forages can be more susceptible to proteolysis during the silage process than grasses, and dairy cows fed these legume silages would excrete a larger quantity of nitrogen in slurry. The objectives of this work were to evaluate the amount of nitrogen excretion in dairy cows fed different protein levels and legume silages with a view to improve the slurry quality as a co-product that can be used as fertilizer. Two double 3 × 3 Latin square trials were carried out in order to study three different protein levels (high, medium, and low) and three different silages (grass, faba bean, and field pea). Dry matter intake, milk production, and composition were not affected by treatments. The excretion of ammonia-N in the urine was almost four times lower in the diet with the lowest protein level. The ammonia-N in the urine was twice as high with the pea silage than faba bean and grass silages. In conclusion, the diet containing 13% of protein meets the protein requirement for lactating cows producing 31 kg daily, with low nitrogen excretion in the urine, and the main pathway for the excretion of surplus nitrogen from legume silages is through urine and the metabolization of pea silage protein goes toward ammonia-N.

64.- VILLAR, A., VÁZQUEZ-GONZÁLEZ, I., VICENTE, F., SALCEDO, G., GONZÁLEZ, L., BOTANA, A., ROYO, L. J., EGUINOA, P., & BUSQUE, J. (2021). Study of the variability in fatty acids and carotenoid profiles: laying the ground for tank milk authentication. *Sustainability, 13*(8), Art. 4506. DOI: 10.3390/su13084506. (OA) [NPF] [Ver online]

Abstract: This study analyzes 174 tank milk samples taken from 89 commercial farms located all along the Cantabrian Coast (Green Spain). Sampling was performed in two periods: autumn 2016 and spring 2017. A survey was carried out for every day of sampling to record the average lactating dairy cow production and its diet composition. For each sample, the fatty acid (FA) profile (49 FA plus its main relationships) and nine fat-soluble antioxidant (FSA) profiles (retinol (vitamin A), α- and Y-tocopherol (vitamin E), all-trans-β-carotene, 9-cis-β-carotene, 13-cis-β-carotene, lutein, zeaxanthin, and β-cryptoxanthin) were determined. The milk production varied between 7.3 and 45.9 liters per cow per day, highlighting the diversity found among production systems. The milk fat content ranged from 2.64% to 4.38% and the protein content from 2.87% to 3.56%. Regarding the fatty acids profile, the percentage of saturated fat varied between 59.95% and 75.99%. The linolenic acid content fluctuated between 0.21 and 1.31 and rumenic acid ranged from 0.20 to 2.47 (g 100 g^{-1} total FA). The most important correlations between diet and milk FA were always related to the content of fresh grass and total forage (which is defined by both fresh and conserved forage derived from fresh grass (GCF)) in the diet. The content of vaccenic acid, linolenic acid, total omega-3, rumenic acid, and total CLA isomers showed the highest correlation with the proportion of fresh grass in the diet. The antioxidant contents were also highly variable, although correlations with dietary components were lower. The highest correlations were between total forage content (fresh grass (FG) plus GCF) and lutein, all-trans-β-

carotene, and 13-cis-β-carotene. Diets without fresh grass had lower omega-3 content, CLA, vaccenic acid, lutein, all-trans-β-carotene, and 13-cis-β-carotene.

65.- YÁNIZ J.L., PALACÍN I., SILVESTRE M.A., HIDALGO C.O., TAMARGO C.& SANTOLARIA, P. (2021.) Ability of the ISAS3Fun method to detect sperm acrosome integrity and its potential to discriminate between high and low field fertility bulls. *Biology,10*, Art. 1135. DOI: 10.3390/biology10111135. (OA) [SRA] [Ver online]

Abstract: The objective of the present study was to investigate whether fertility differences in bulls are reflected in variations of sperm quality when analysing only one ejaculate per male. Two experiments were performed. In the first experiment, frozen semen samples from 20 adult bulls were tested; 10 bulls had high field fertility and 10 bulls had low field fertility. Analyses of sperm motility, membrane integrity, and membrane–acrosome integrity with the ISAS3Fun method were performed. Sperm morphometry of the fluorescence sperm subpopulations obtained with the ISAS3Fun method was also analysed. Significant differences between high- and low-fertility groups were only found with the ISAS3Fun technique, specifically in sperm acrosome integrity, the proportion of spermatozoa with an intact acrosome and damaged membrane, and in sperm head width of spermatozoa with intact structures. Discriminant analyses allowed us to correctly classify 90% of sperm samples in their fertility group using sperm quality parameters. Given that only the results obtained with the ISAS3Fun technique were related to bull fertility, we performed a second experiment aimed to validate the efficacy of this technique to detect the acrosomal integrity of bull spermatozoa, comparing them with the conventional FITC-PNA/propidium iodide (PNA/PI) combination under capacitating conditions. The results indicated that the ISAS3Fun combination provided an accurate assessment of both viability and acrosomal integrity for ejaculated spermatozoa, while the PNA/PI combination underestimated the extension of acrosomal damage due to false negatives. It was concluded that the simultaneous assessment of sperm plasma membranes and acrosome integrity with the ISAS3Fun method is precise and seems to have a greater potential to discriminate between high- and low-fertility bulls than more conventional in vitro sperm quality tests.

66.- ZAMUZ, S., GARCÍA-TORRES, S., CABEZA DE VACA, M., TEJERINA, D., ORTIZ, A., OLIVÁN. M., SIERRA, V., DIÑEIRO, Y., SENTANDREU, M.A., LÓPEZ-PEDROUSO, M., PURRIÑOS, L., LORENZO, J.M., & FRANCO. D. (2021ª). Impacto del modo de transporte al matadero (mezcla o no mezcla con animales extraños) de terneros de las razas Asturiana de los Valles y Retinta en los atributos físico-químicos y organolépticos de carne madurada. Aceptabilidad y preferencia de los consumidores. *ITEA-Información Técnica Económica Agraria.* DOI: 10.12706/itea.2021.021. (OA) [SPA] [Ver online]

Abstract: As ageing process is one of the main factors influencing the organoleptic characteristics of meat and its consumer acceptability, the objective of this research was to study the organoleptic attributes variation of meat from Asturiana de los Valles and Retinta breeds at two times of the ageing process (7 and 14 days) under different pre-slaughter transport conditions (mixing and non-mixing with unfamiliar animals). A descriptive sensory analysis was carried out with a trained panel and the acceptability and preference for ageing meat of these two breeds were analysed with 75 consumers, as well as a study on the factors that influence the purchase decision to this type of meat. The breed and mixing with unfamiliar animals before slaughter affected

significantly at instrumental tenderness. The intensity of the attributes related to the texture showed significant differences among studied samples at 7 days of ageing, with an increase in tenderness, juiciness and fibrousness while at 14 days of ageing, odour, beef flavour, liver flavour and smoothness also showed significant differences. The results indicated that the organoleptic attributes changed in intensity during ageing time, therefore, we can conclude that the attributes of tenderness, juiciness, smoothness fibrousness, odour, beef flavour, liver flavour showed significant differences among samples, could be considered sensory indicators of ageing. The type of pre-slaughter transport (with or without mixing with unknown animals) also influenced the intensity of the sensory attributes, but less markedly than the breed effect. It was also observed that consumers prefer the meat of the Asturiana de los Valles breed aged for 14 days, despite among the factors that most influence the meat purchase decision, the ageing time was not present. Regarding purchasing habits, a relationship was observed between the meat purchase format and sale point. Overall, it is necessary to promote greater knowledge among consumers about the importance of ageing in beef quality.

ÍNDICES

A) ÍNDICE ALFABÉTICO DE AUTORES DEL SERIDA

- SIERRA SÁNCHEZ, Verónica: (36), (61), (66)
- SUÁREZ VALLES, María Belén (TA): *(11), (51), (54), (56)*
- TAMARGO DE MIGUEL, Carolina (SRA): *(9), (42), (59), (65)*
- VICENTE MAINAR, Fernando (NPF): *(5), (63), (64)*

B) ÍNDICE ALFABÉTICO DE TÍTULOS DE REVISTAS CON INDICACIÓN DE SU FACTOR DE IMPACTO (FI)

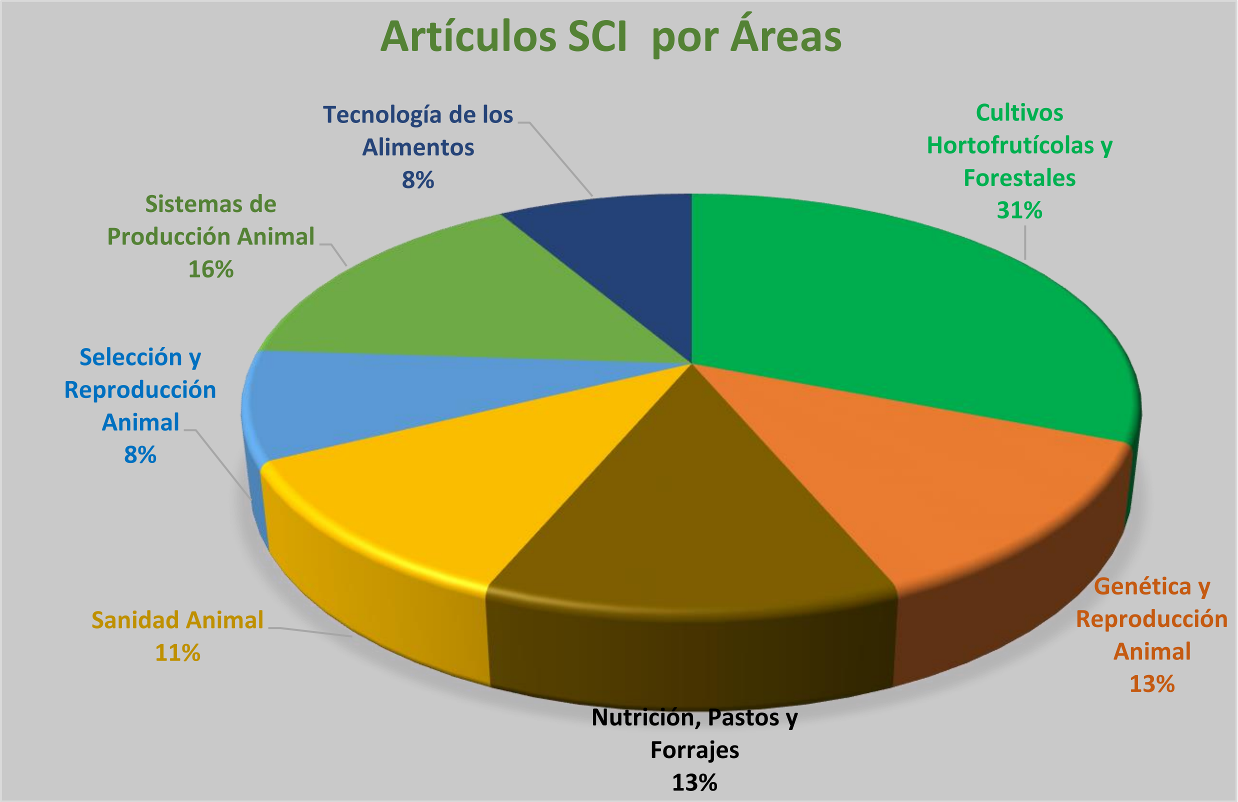
	Nº artículos	CUARTIL	FI 2020	Referencia	Área
Agriculture Ecosystems and Environment	1	Q1	5,56	*(40)*	CHF
Agronomy Basel	2	Q1	3,41	*(18)*, (46)**	CHF(2)
Animals	8	Q1	2,75	*(1)*, (3)*, (7)*, (8)*, (9)*, (21)*, (23)*, (29)* (47)*, (63)**	CHF, GRA(2) SA(3) SPA(2) SRA NPF
Ardeola International Journal of Ornithology	1	Q2	1,47	*(20)**	CHF
Basic and Applied Ecology	1	Q2	3,41	*(39)**	CHF
Beverages	1	Q3	0,54	*(51)**	TA
Biology Basel	2	Q1	5,07	*(43)*, (65)**	SRA(2)
Bmc Plant Biology	1	Q1	4,21	*(26)+*	CHF
Carbohydrate Polymers	1	Q1	9,38	*(10)**	CHF
Crop Protection	1	Q2	2,57	*(44)*	CHF
Ecological Applications: A Publication of the Ecological Society of America	1	Q1	4,65	*(30)**	CHF
Ecology	1	Q1	5,49	*(2)**	CHF
European Food Research and Technology	1	Q2	2,99	*(55)*	TA
European Journal of Agronomy	1	Q1	5,12	*(16)*	CHF
Foods	3	Q2	4,35	*(36)*, (54)*, (61)**	SPA(2) (TA-CHF)
Frontiers in Cell and Developmental Biology	2	Q2	6,68	*(14)*, (53)**	SPA SRA
Frontiers in Cellular and Infection Microbiology	1	Q1	5,29	*(28)**	SPA
Frontiers in Plant Science	2	Q1	5,75	*(11)*, (22)**	(CHF-TA) CHF
Frontiers in Veterinary Science	1	Q1	3,41	*(52)**	SA
Gene	1	Q2	3,68	*(38)*	GRA
Genetics Selection Evolution	1	Q1	4,29	*(19)**	GRA
Global Ecology and Conservation	1	Q1	3,38	*(50)**	CHF
Insects	1	Q1	2,76	*(45)**	CHF
Itea: Información Técnica Económica Agraria	2	Q4	0,54	*(15)*, (66)**	NPF SPA
Journal of Agricultural and Food Chemistry	1	Q1	5,27	*(60)**	SPA
Journal of Animal and Plant Sciences Japs	1	Q4	0,49	*(4)**	NPF
Journal of Animal Breeding and Genetics	1	Q2	2,38	*(37)*	GRA
Journal of Animal Science and Biotechnology	1	Q1	5,03	*(42)**	SRA
Journal of Applied Ecology	1	Q1	6,52	*(57)**	CHF

Journal of Proteomics	1	Q2	4,04	*(25)*	SPA
Journal of the Science of Food and Agriculture	1	Q1	3,63	*(24)**	SPA
Metabolites	2	Q2	4,93	*(31),(34)**	GRA
Molecules	1	Q2	4,41	*(56)**	TA
Pharmaceutics	1	Q1	6,32	*(62)**	NPF
Plant Disease	1	Q1	4,43	*(13)**	(CHF-TA)
Plant Journal	1	Q1	6,48	*(6)**	CHF
Reproduction, Fertility and Development	2	Q4	2,31	*(32)*, (33)**	GRA
Research in Veterinary Science	1	Q1	2,53	*(59)*	SRA
Scientia Horticulturae	1	Q1	3,46	*(17)*	CHF
Scientific Reports	1	Q1	4,38	*(12)**	SA
Small Ruminants Research	2	Q3	1,61	*(35), (58)*	SPA, NPF
Sustainability	2	Q2	3,25	*(5)*, (64)**	NPF(2)
Theoretical and Applied Genetics	1	Q1	5,69	*(27)*	CHF
Transboundary and Emerging Diseases	2	Q1	5,00	*(48), (49)**	(SA-NPF)(2)
Vaccine	1	Q3	3,64	*(41)*	SA

ARTÍCULOS POR ÁREAS DE INVESTIGACIÓN DEL SERIDA

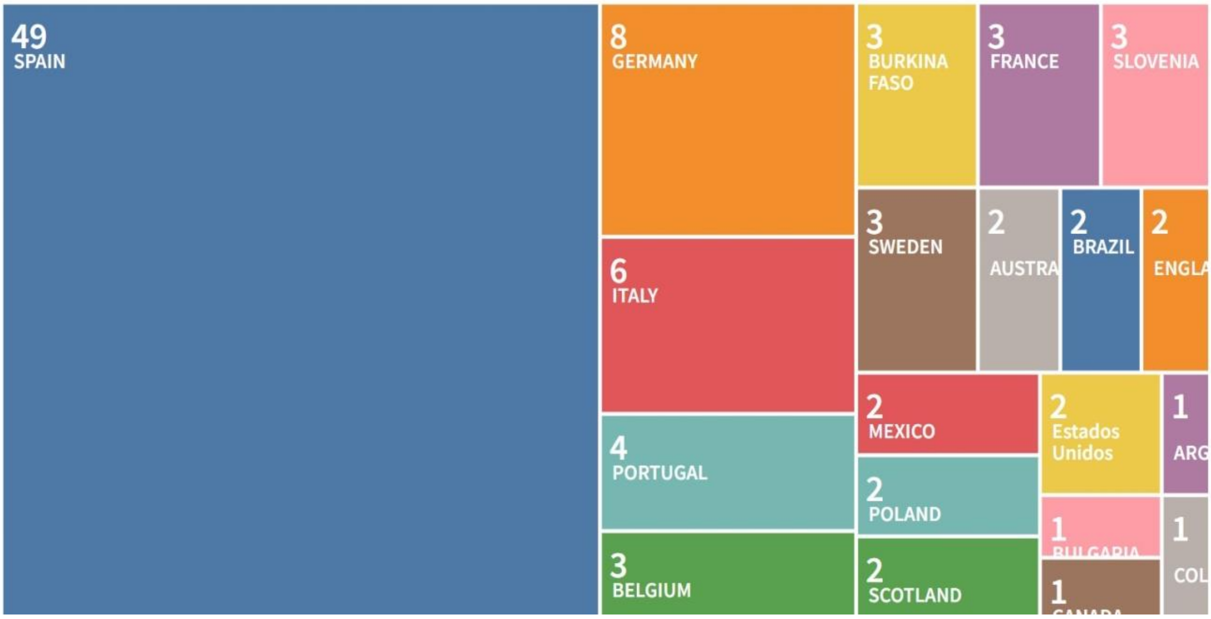
- ÁREA DE CULTIVOS HORTOFRUTÍCOLAS Y FORESTALES: *(1), (2), (6), (10), (11), (13), (16), (17), (18), (20), (22), (26), (27), (30), (39), (40), (44), (45), (46), (50), (54), (57).*
- ÁREA DE GENÉTICA Y REPRODUCCIÓN ANIMAL: *(3), (19), (31), (32), (33), (34), (37), (38), (47).*
- ÁREA DE NUTRICIÓN, PASTOS Y FORRAJES: *(4), (5), (15), (48), (49), (58), (62), (63), (64).*
- ÁREA DE SANIDAD ANIMAL: *(7), (8), (12), (21), (41), (48), (49), (52).*
- ÁREA DE SELECCIÓN Y REPRODUCCIÓN ANIMAL: *(9), (42), (43), (53), (59, (65).*
- ÁREA DE SISTEMAS DE PRODUCCIÓN ANIMAL: *(14), (23), (24), (25), (28), (29), (35), (36), (60), (61), (66).*
- ÁREA DE TECNOLOGÍA DE LOS ALIMENTOS*: (11), (13), (51), (54), (55), (56).*

Figura 1: Artículos por áreas de investigación del SERIDA



Fuente: Elaboración propia

Figura 2: Número de artículos realizados en colaboración con otros países



Fuente: Web of Science

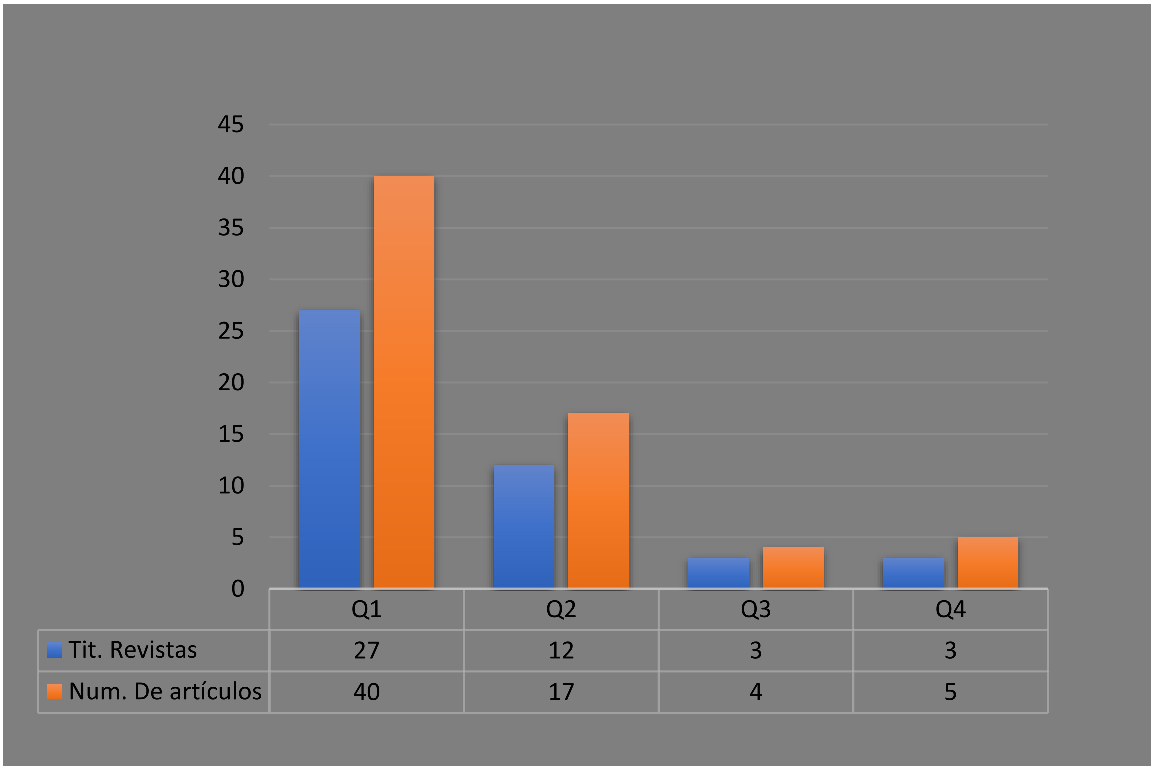
Figura 3: Número de artículos realizados en colaboración con otras instituciones

Fuente: Web of Science

Figura 4: Número de artículos por título de revista

Fuente: Web of Science

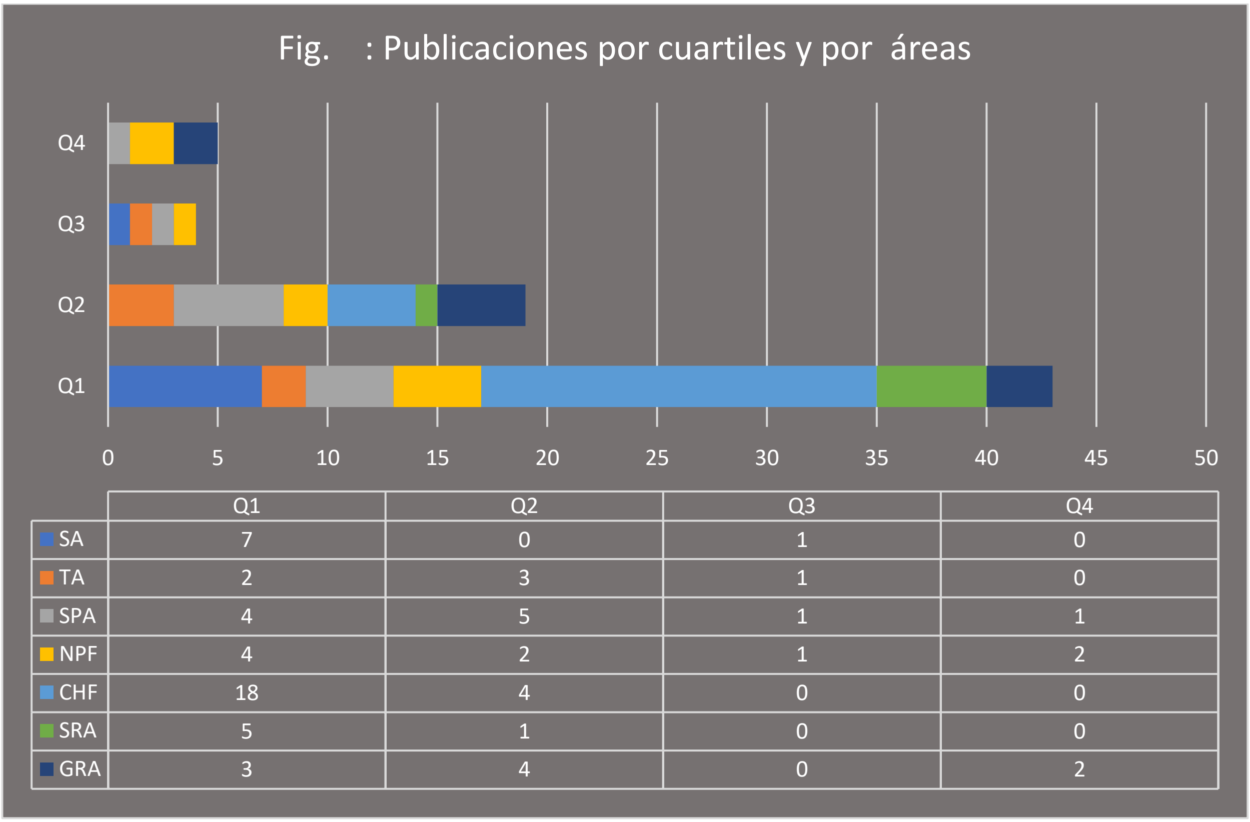
Figura 5: Cuartiles de las revistas en las que aparecen publicados los artículos del SERIDA



	Q1	Q2	Q3	Q4
🟦 Tit. Revistas	27	12	3	3
🟧 Num. De artículos	40	17	4	5

Fuente: Elaboración propia

Figura 6: Cuartiles de las revistas en las que aparecen publicados los artículos del SERIDA según las distintas áreas de investigación



	Q1	Q2	Q3	Q4
🟦 SA	7	0	1	0
🟧 TA	2	3	1	0
⬜ SPA	4	5	1	1
🟨 NPF	4	2	1	2
🟦 CHF	18	4	0	0
🟩 SRA	5	1	0	0
🟦 GRA	3	4	0	2

Fuente: Elaboración propia