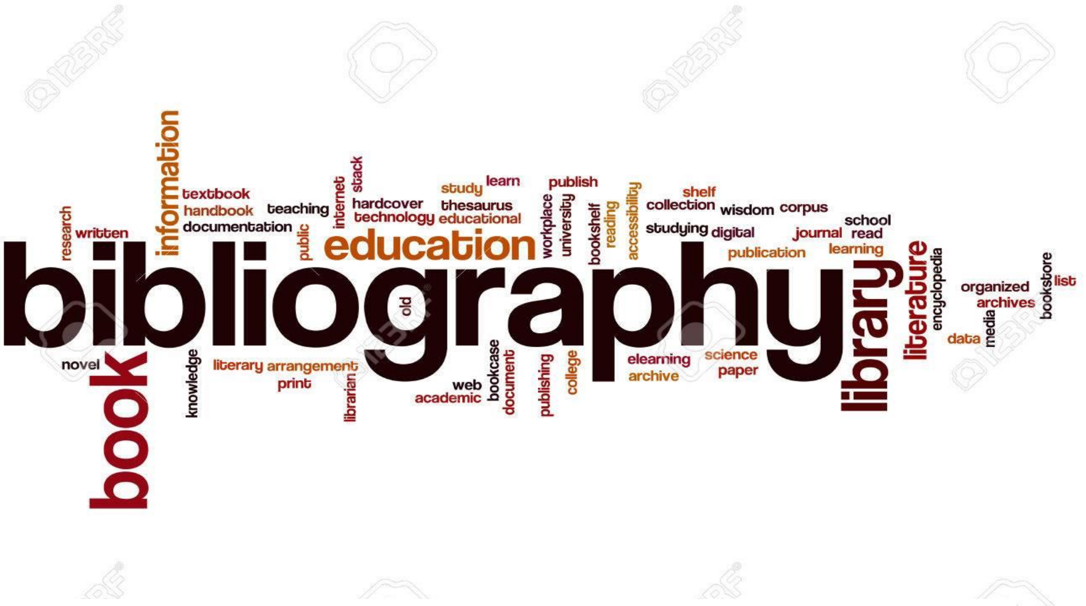

Bibliografía Científica

Anexo 2023

SERIDA
Servicio Regional de Investigación
y Desarrollo Agroalimentario

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[Abril 2024]

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 2023 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 2023, aceptándose además para su inclusión en la bibliografía, aquellos registros que tuvieran fecha de "acceso anticipado" en 2023, 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 44 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 35 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), las instituciones con las que se ha colaborado (Figura 2), los títulos de las revistas en las que más se ha publicado (Figura 3), y la importancia de la revista en la que aparecen publicados los artículos (Figuras 4 y 5).

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).

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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.- **ABOU EL QASSIM, L.**, MARTÍNEZ, B., RODRÍGUEZ, A., DÁVALOS, A., LÓPEZ DE LAS HAZAS, M.-C., **MENÉNDEZ MIRANDA, M.**, & **ROYO, L. J**. (2023). Effects of cow's milk processing on MicroRNA levels. *Foods*, *12*(15), art. 2950. DOI: 10.3390/foods12152950A. (OA) [NPF] [Ver online]

Abstract: MicroRNAs (miRNAs) regulate gene expression and might resist adverse physicochemical conditions, which makes them potential biomarkers. They are being investigated as biomarkers of dairy production systems, based on the variations in their levels in raw milk depending on animal diet and management. Whether miRNA levels can serve as biomarkers for dairy products remains unclear, since technological or culinary treatments, such as fermentation, may alter their levels. Here, 10 cow dairy farms were sampled in Asturias (north-west Spain) and milk samples were subjected to microwave heating or used to produce yogurt or cheese. Total RNA was isolated from raw milk and three derived products, and levels of seven miRNAs, selected based on previous studies as possible milk production system biomarkers, were assessed by RT-qPCR. The treatments decreased levels of all miRNAs to some extent. These results also imply that cheesemaking increases the concentration of miRNAs in this product; raw milk and cheese supposedly may provide similar concentrations of miRNAs, higher than those of yogurt and microwaved milk. They also indicate that the content of certain miRNAs in raw milk cannot necessarily be extrapolated to other dairy products.

2.- ALONSO-HEARN, M., BALLESTEROS, A., **NAVARRO, A.,** BADÍA-BRINGUÉ, G**.,** & **CASAIS, R.** (2023). Lateral-flow assays for bovine paratuberculosis diagnosis [Review] *Frontiers in Veterinary Science*, *10*, art. 1257488. DOI: 10.3389/fvets.2023.1257488 (OA) [SA] [Ver online]

Abstract: *Mycobacterium avium* subsp. *paratuberculosis* (MAP) causes bovine paratuberculosis (PTB). PTB is responsible for significant economic losses in dairy herds around the word. PTB control programs that rely on testing and culling of test-positive cows have been developed. Current diagnostics, such as ELISA for detecting MAP antibodies in serum samples and PCR detecting MAP DNA in feces, have inadequate sensitivity for detecting subclinical animals. Innovative "omics" technologies such as next-generation sequencing (NGS) technology-based RNA-sequencing (RNA-Seq), proteomics and metabolomics can be used to find host biomarkers. The discovered biomarkers (RNA, microRNAs, proteins, metabolites) can then be used to develop new and more sensitive approaches for PTB diagnosis. Traditional approaches for measuring host antibodies and biomarkers, such as ELISAs, northern blotting, quantitative reverse-transcriptase polymerase chain reaction (RT-qPCR), cDNA microarrays, and mass spectrometry are time-consuming, expensive, and sometimes exhibit poor sensitivity. With the rapid development of nanotechnology, low-cost monitoring devices for measuring antibodies against MAP proteins in point-of-care (POC) settings have been developed. Lateral flow assays (LFAs), in particular, are thought to be appropriate for the on-site detection of antibodies to MAP antigens and/or host biomarkers. This review aims to summarize LFAs that have recently been developed to accurately detect antibodies against MAP antigens, as well as the benefits that host biomarkers linked with MAP infection give to PTB diagnosis. The identification of these novel biomarkers could be the basis for the development of new LFAs. The dairy industry and producers are likely to benefit from reliable and rapid technologies capable of detecting MAP infection *in situ* to establish a quick and sensitive PTB diagnosis.

3.- ÁLVAREZ, J.M., RODRÍGUEZ, S.M., **FUENTE-MAQUEDA, F.**, **FEITO, I.**, ORDÁS, R.J., & CUESTA, C. (2023). Identification of candidate genes involved in bud growth in *Pinus pinaster* through knowledge transfer from *Arabidopsis thaliana* models. *Forests*, *14*, Art. 1765. DOI: 10.3390/f14091765 (OA) [CHF] [Ver online]

Abstract: *Pinus pinaster* is a plant species of great ecological and economic importance. Understanding the underlying molecular mechanisms that govern the growth and branching of P. pinaster is crucial for enhancing wood production and improving product quality. In this study, we describe a simple methodology that enables the discovery of candidate genes in *Pinus pinaster* by transferring existing knowledge from model species like *Arabidopsis thaliana* and focusing on factors involved in plant growth, including hormonal and non-hormonal pathways. Through comparative analysis, we investigated the main genes associated with these growth-related factors in A. thaliana. Subsequently, we identified putative homologous sequences in *P. pinaster* and assessed the conservation of their functional domains. In this manner, we can exclude sequences that, despite displaying high homology, lack functional domains. Finally, we took an initial approach to their validation by examining the expression levels of these genes in P. pinaster trees exhibiting contrasting growth patterns. This methodology allowed the identification of 26 candidate genes in *P. pinaster*. Our findings revealed differential expression patterns of key genes, such as *NCED3, NRT1.2, PIN1, PP2A, ARF7, MAX1, MAX2, GID1, AHK4, AHP1*, and *STP1*, in relation to the different growth patterns analyzed. This study provides a methodological foundation for further exploration of these genes involved in the growth and branching processes of *P. pinaster*. This will contribute to the understanding of this important tree species and open new avenues for enhancing its utilization in sustainable forestry practices.

4.- **ARIAS, K.D.**, GUTIÉRREZ, J.P., **FERNÁNDEZ, I.**, **ÁLVAREZ, I.** & **GOYACHE, F.** (2023) Approaching autozygosity in a small pedigree of Gochu Asturcelta pigs. *Genetics Selection Evolution, 55*(1), art.74. DOI: 10.1186/s12711-023-00846-7 (OA) [GRA] [Ver online]

Abstract: Background: In spite of the availability of single nucleotide polymorphism (SNP) array data, differentiation between observed homozygosity and that caused by mating between relatives (autozygosity) introduces major difficulties. Homozygosity estimators show large variation due to different causes, namely, Mendelian sampling, population structure, and differences among chromosomes. Therefore, the ascertainment of how inbreeding is reflected in the genome is still an issue. The aim of this research was to study the usefulness of genomic information for the assessment of genetic diversity in the highly endangered Gochu Asturcelta pig breed. Pedigree depth varied from 0 (founders) to 4 equivalent discrete generations (*t*). Four homozygosity parameters (runs of homozygosity, F_{ROH}; heterozygosity-rich regions, F_{HRR}; Li and Horvitz's, F_{LH}; and Yang and colleague's F_{YAN}) were computed for each individual, adjusted for the variability in the base population (BP; six individuals) and further jackknifed over autosomes. Individual increases in homozygosity (depending on *t*) and increases in pairwise homozygosity (i.e., increase in the parents' mean) were computed for each individual in the pedigree, and effective population size (N_e) was computed for five subpopulations (cohorts). Genealogical parameters (individual inbreeding, individual increase in inbreeding, and N_e) were used for comparisons.- Results: The mean *F* was 0.120 ± 0.074 and the mean BP-adjusted homozygosity ranged from 0.099 ± 0.081 (F_{LH}) to

0.152 ± 0.075 (F_{YAN}). After jackknifing, the mean values were slightly lower. The increase in pairwise homozygosity tended to be twofold higher than the corresponding individual increase in homozygosity values. When compared with genealogical estimates, estimates of N_e obtained using F_{YAN} tended to have low root-mean-squared errors. However, N_e estimates based on increases in pairwise homozygosity using both F_{ROH} and F_{HRR} estimates of genomic inbreeding had lower root-mean-squared errors.-Conclusions: Parameters characterizing homozygosity may not accurately depict losses of variability in small populations in which breeding policy prohibits matings between close relatives. After BP adjustment, the performance of F_{ROH} and F_{HRR} was highly consistent. Assuming that an increase in homozygosity depends only on pedigree depth can lead to underestimating it in populations with shallow pedigrees. An increase in pairwise homozygosity computed from either F_{ROH} or F_{HRR} is a promising approach for characterizing autozygosity.

5.- **ARIAS, K.D.**, GUTIÉRREZ, J.P., **FERNÁNDEZ, I**., **ÁLVAREZ, I**. & **GOYACHE, F.** (2023) Copy number variation regions differing in segregation patterns span different sets of genes. *Animals*, *13*, 2351. DOI: 10.3390/ani13142351. (OA) [GRA] [Ver online]

Abstract: Copy number variations regions (CNVRs) can be classified either as segregating, when found in both parents, and offspring, or non-segregating. A total of 65 segregating and 31 non-segregating CNVRs identified in at least 10 individuals within a dense pedigree of the Gochu Asturcelta pig breed was subjected to enrichment and functional annotation analyses to ascertain their functional independence and importance. Enrichment analyses allowed us to annotate 1018 and 351 candidate genes within the bounds of the segregating and non-segregating CNVRs, respectively. The information retrieved suggested that the candidate genes spanned by segregating and non-segregating CNVRs were functionally independent. Functional annotation analyses allowed us to identify nine different significantly enriched functional annotation clusters (ACs) in segregating CNVR candidate genes mainly involved in immunity and regulation of the cell cycle. Up to five significantly enriched ACs, mainly involved in reproduction and meat quality, were identified in non-segregating CNVRs. The current analysis fits with previous reports suggesting that segregating CNVRs would explain performance at the population level, whereas non-segregating CNVRs could explain between-individuals differences in performance.

6.- **ARIAS, K.D**., LEE, H., BOZZI, R., **ÁLVAREZ, I.**, GUTIÉRREZ, J. P., **FERNÁNDEZ, I**., MENÉNDEZ, J., BEJA-PEREIRA, A., & **GOYACHE, F**. (2023^{a}- 2024^{p}). Ascertaining the genetic background of the Celtic-Iberian pig strain: A signatures of selection approach. *Journal of Animal Breeding and Genetics*, *141*(1), 96-112. DOI: 10.1111/jbg.1282 (OA) [GRA] [Ver online]

Abstract: Celtic-Iberian pig breeds were majority in Spain and Portugal until the first half of the 20th century. In the 1990s, they were nearly extinct as a result of the introduction of foreign improved pig breeds. Despite its historical importance, the genetic background of the Celtic-Iberian pig strain is poorly documented. In this study, we have identified genomic regions that might contain signatures of selection peculiar of the Celtic-Iberian genetic lineage. A total of 153 DNA samples of Celtic-Iberian pigs (Spanish Gochu Asturcelta and Portuguese Bisara breeds), Iberian pigs (Spanish Iberian and Portuguese Alentejano breeds), Cinta Senese pig, Korean local pig and Cosmopolitan pig

(Hampshire, Landrace and Large White individuals) were analysed. A pairwise-comparison approach was applied: the Gochu Asturcelta and the Bisara samples as test populations and the five other pig populations as reference populations. Three different statistics (*XP-EHH*, *FST* and *DeltaDAF*) were computed on each comparison. Strict criteria were used to identify selection sweeps in order to reduce the noise brought on by the Gochu Asturcelta and Bisara breeds' severe population bottlenecks. Within test population, SNPs used to construct potential candidate genomic areas under selection were only considered if they were identified in four of ten two-by-two pairwise comparisons and in at least two of three statistics. Genomic regions under selection constructed within test population were subsequently overlapped to construct candidate regions under selection putatively unique to the Celtic-Iberian pig strain. These genomic regions were finally used for enrichment analyses. A total of 39 candidate regions, mainly located on SSC5 and SSC9 and covering 3130.5kb, were identified and could be considered representative of the ancient genomic background of the Celtic-Iberian strain. Enrichment analysis allowed to identify a total of seven candidate genes (*NOL12, LGALS1, PDXP, SH3BP1, GGA1, WIF1,* and *LYPD*6). Other studies reported that the *WIF1* gene is associated with ear size, one of the characteristic traits of the Celtic-Iberian pig strain. The function of the other candidate genes could be related to reproduction, adaptation and immunity traits, indirectly fitting with the rusticity of a non-improved pig strain traditionally exploited under semi-extensive conditions.

7.- BADIA-BRINGUÉ, G., CANIVE, M., FERNANDEZ-JIMENEZ, N. LAVÍN, J. L., **CASAIS, R**., **BLANCO-VÁZQUEZ, C**., VÁZQUEZ, P., FERNÁNDEZ, A., BILBAO, J.R., GARRIDO, J.M., JUSTE, R.A., GONZÁLEZ-RECIO, & ALONSO-HEARN, M. (2023). Summary-data based Mendelian randomization identifies gene expression regulatory polymorphisms associated with bovine paratuberculosis by modulation of the *nuclear factor Kappa β (NF-κß)*-mediated inflammatory response*, BMC Genomics 24*(1), art. 605. DOI: 10.1186/s12864-023-09710-w. (OA) [SA] [Ver online]

Abstract: Genome-wide association studies (GWAS) have identified host genetic variants associated with paratuberculosis (PTB) susceptibility. Most of the GWAS-identified SNPs are in non-coding regions. Connecting these non-coding variants and downstream affected genes is a challenge and, up to date, only a few functional mutations or expression quantitative loci (cis-eQTLs) associated with PTB susceptibility have been identified. In the current study, the associations between imputed whole-genome sequence genotypes and whole RNA-Sequencing data from peripheral blood (PB) and ileocecal valve (ICV) samples of Spanish Holstein cows ($N = 16$) were analyzed with *TensorQTL*. This approach allowed the identification of 88 and 37 cis-eQTLs regulating the expression levels of 90 and 37 genes in PB and ICV samples, respectively (False discorey rate, $\text{FDR} \leq 0.05$). Next, we applied summary-based data Mendelian randomization (SMR) to integrate the *cis*-eQTL dataset with GWAS data obtained from a cohort of 813 culled cattle that were classified according to the presence or absence of PTB-associated histopathological lesions in gut tissues. After multiple testing corrections ($\text{FDR} \leq 0.05$), we identified two novel cis-eQTLs affecting the expression of the *early growth response factor 4* (*EGR4*) and the bovine *neuroblastoma breakpoint family member 6-like protein* isoform 2 (*MGC134040*) that showed pleiotropic associations with the presence of multifocal and diffuse lesions in gut tissues; $P = 0.002$ and $P = 0.017$, respectively. While *EGR4* acts as a brake on T-cell proliferation and cytokine production through interaction with the *nuclear factor Kappa β* (*NF-κß), MGC134040* is a target gene of *NF-κß*. Our findings provide a better understanding of the genetic factors influencing PTB outcomes, confirm that the multifocal lesions are localized/confined lesions that have different underlying host genetics than the diffuse lesions, and highlight regulatory SNPs and regulated-gene targets to design future functional studies.

8.- BALSEIRO, A., HERRERO, G., **ROYO, L J**., ARMENTEROS, J, A., ALTONAGA, J.R., MONASTERIO, J.M., BALSERA, R., POOL, R.V., GARCÍA, J.F., & PIS, J.A. (2023). Hypertrophic osteopathy in a common dolphin (*Delphinus delphis*) with concurrent pulmonary *Halocercus delphini* infestation, *Heliyon* 9(6), art. e17011. DOI: 10.1016/j.heliyon.2023.e17011. (OA) [NPF] [Ver online]

Abstract: Dolphins are marine mammals that often live in coastal habitats. Common causes of severe skeletal disorders among wild dolphins are congenital vertebral anomalities, collisions with sea vessels, trauma, hunting-related injury, infectious diseases, environmental pollution, and tumors. A free-ranging male, 3-year-old common dolphin (*Delphinus delphis*) was found dead in the coast of Asturias in northern Spain. Postmortem examination revealed lordosis in the caudal vertebral column, while X-ray imaging and computer tomography showed well-organized palisade-like periosteal proliferation, appearing as florid-like accretions, along the spinous apophysis of 26 lumbar-caudal vertebrae. The transverse apophysis was affected on only a few caudal vertebrae. The cortical layer remained intact. Histology of vertebra tissue showed periosteal proliferation of cancellous bone. The animal was diagnosed with hypertrophic osteopathy. The lungs showed diffuse parasitic granulomatous bronchointerstitial pneumonia caused by *Halocercus delphini*, consolidation of the pulmonary tissue, congestion, and alveolar edema. The animal was also afflicted by parasitic granulomatous gastritis caused by *Anisakis simplex sensu lato* and tattoo skin disease. The dolphin suffered from hypertrophic osteopathy associated with pulmonary *Halocercus delphini* infestation. This syndrome, known as hypertrophic pulmonary osteopathy, has been described in diverse terrestrial mammals, including domestic animals, wildlife and humans, but not in dolphins. This case reports the first description of hypertrophic osteopathy associated to a pulmonary disorder in dolphin, and it provides insights into factors that can induce column malformation in dolphins, suggesting the importance of taking thoracic lesions into account during differential diagnosis.

9.- BELLUCCI, E., BENAZZO, A., XU, C., BITOCCHI, E., RODRIGUEZ, M., ALSEEKH, S., DI VITTORI, V., GIOIA, T., NEUMANN, K., CORTINOVIS, G., FRASCARELLI, G., MURUBE, E., TRUCCHI, E., NANNI, L., ARIANI, A., LOGOZZO, G., SHIN, J.H., LIU, C., JIANG, L., **FERREIRA, J.J**., **CAMPA, A**., ATTENE, G., MORRELL, P., BERTORELLE, G., GRANER, A., GEPTS, P., FERNIE, A., JACKSON, S., & PAPA, R. (2023) Selection and adaptive introgression guided the complex evolutionary history of the European common bean. *Nature Communications, 14*(1), art. 1908. DOI: 10.1038/s41467-023-37332-z (OA) [CHF] [Ver online]

Abstract: Domesticated crops have been disseminated by humans over vast geographic areas. Common bean (*Phaseolus vulgaris L*.) was introduced in Europe after 1492. Here, by combining whole-genome profiling, metabolic fingerprinting and phenotypic characterisation, we show that the first common bean cultigens successfully introduced into Europe were of Andean origin, after Francisco Pizarro's expedition to northern Peru in 1529. We reveal that hybridisation, selection and recombination have shaped the genomic diversity of the European common bean in parallel with political constraints. There is clear evidence of adaptive introgression into the Mesoamerican-derived European genotypes, with 44 Andean introgressed genomic segments shared by more than 90% of European accessions and distributed across all chromosomes except PvChr11. Genomic scans for signatures of selection highlight the role of genes relevant to flowering and environmental adaptation, suggesting that introgression has been crucial for the dissemination of this tropical crop to the temperate regions of Europe. Common bean has two distinct domestication centers in Mesoamerica and in the Andes. The authors show that the Andean is the first gene pool successfully introduced in Europe and identify signature of pervasive introgression among gene pools and of selection for flowering underlying adaptation.

10.- **CAAMAÑO, J.N.**, SANTIAGO, J., MARTÍNEZ, F., **TAMARGO, C.**, SALMAN, A., **FERNÁNDEZ, Á.**, **MERINO, M.J.**, LACALLE, E., TOLEDANO, A. & **HIDALGO, C.O.** (2023) Use of the flavonoid taxifolin for sperm cryopreservation from the threatened Bermeya goat breed. *Theriogenology*, *206*, 18-27. DOI: 10.1016/j.theriogenology.2023.05.004. [SRA] [Ver online]

Abstract: Taxifolin is a plant flavonoid effective as an antioxidant. This study aimed to assess the effect of adding taxifolin to the semen extender during the cooling period before freezing on the overall post-thawing sperm variables of Bermeya goats. In the first experiment, a dose-response experiment was performed with four experimental groups: Control, 10, 50, and 100mug/ml of taxifolin using semen from 8 Bermeya males. In the second experiment, semen from 7 Bermeya bucks was collected and extended at 20°C using a Tris-citric acid-glucose medium supplemented with different concentrations of taxifolin and glutathione (GSH): control, 5muM taxifolin, 1mM GSH, and both antioxidants. In both experiments, two straws per buck were thawed in a water bath (37°C, 30s), pooled, and incubated at 38°C. Motility (CASA) was assessed at 0, 2, and 5h, and sperm physiology was assessed at 0 and 5h by flow cytometry (viability, intact acrosome membrane, mitochondria membrane potential, capacitation, intracellular reactive oxygen species -ROS-, mitochondrial superoxide, and chromatin status). In experiment 2, an artificial insemination trial (AI) was included with 29 goats for testing the taxifolin 5-muM treatment on fertility. Data were analyzed with the R statistical environment using linear mixed-effects models. In experiment 1 and compared to the control, T10 increased progressive motility ($P<0.001$) but taxifolin decreased total and progressive motility at higher concentrations ($P<0.001$), both post-thawing and after the incubation. Viability decreased post-thawing in the three concentrations ($P<0.001$). Cytoplasmic ROS decreased at 0 and 5h at T10 ($P=0.049$), and all doses decreased mitochondrial superoxide post-thawing ($P=0.024$). In experiment 2, 5muM taxifolin or 1mM GSH (alone or combined) increased total and progressive motility vs. the control ($P<0.01$), and taxifolin increased kinematic parameters such as VCL, ALH, and DNC ($P<0.05$). Viability was not affected by taxifolin in this experiment. Both antioxidants did not significantly affect other sperm physiology parameters. The incubation significantly affected all the parameters ($P<0.004$), overall decreasing sperm quality. Fertility after artificial insemination with doses supplemented with 5muM taxifolin was 76.9% (10/13), not significantly different from the control group (69.2%, 9/13). In conclusion, taxifolin showed a lack of toxicity in the low micromolar range and could benefit goat semen cryopreservation.

11.- **CAMPA, A., RODRÍGUEZ MADRERA, R., JURADO, M.**, **GARCÍA-FERNÁNDEZ, C.**, **SUÁREZ VALLES, B., & FERREIRA, J.J.** (2023). Genome-wide association study for the extractable phenolic profile and coat color of common bean seeds (*Phaseolus vulgaris* L.). *BMC Plant Biology 23*(1), 158-174. DOI: 10.1186/s12870-023-04177-z (OA) [CHF]

[TA] [Ver online]

Abstract: A large variation in seed coat colors and seed phenolic metabolites is present in common bean (*Phaseolus vulgaris L*.). The study of the relationships between seed coat color phenotype and the phenolic profile is an important step in the elucidation of the gene network involved in the phenylpropanoid biosynthetic pathway. However, this relationship is still poorly understood in this species. RESULTS: A genome-wide association study (GWAS) was used to investigate the genomic regions associated with the synthesis of 10 flavonoids (5 anthocyanins and 5 flavonols) and with 10 seed coat color traits using a set of 308 common bean lines of the Spanish Diversity Panel (SDP) which have been genotyped with 11,763 SNP markers.. A total of 31 significant SNP-trait associations (QTNs) were identified, grouped in 20 chromosome regions: 6 for phenolic metabolites on chromosomes Pv01, Pv02, Pv04, Pv08, and Pv09, 13 for seed coat color on chromosomes Pv01, Pv02, Pv06, Pv07, and Pv10, and 1 including both types of traits located on chromosome Pv08. In all, 58 candidate genes underlying these regions have been proposed, 31 of them previously described in the phenylpropanoid pathway in common bean, and 27 of them newly proposed in this work based on the association study and their homology with Arabidopsis anthocyanin genes.; CONCLUSIONS: Chromosome Pv08 was identified as the main chromosome involved in the phenylpropanoid pathway and in consequence in the common bean seed pigmentation, with three independent chromosome regions identified, Phe/C_Pv08(2.7) (expanding from 2.71 to 4.04 Mbp), C_Pv08(5.8) (5.89-6.59 Mbp), and Phe_Pv08(62.5) (62.58 to 63.28 Mbp). Candidate genes previously proposed by other authors for the color genes *V* and *P* were validated in this GWAS. Candidate genes have been tentatively proposed from this study for color genes B and Rk on Pv02, *Asp* on Pv07, and complex *C* on Pv08. These results help to clarify the complex network of genes involved in the genetic control of phenolic compounds and seed color in common bean and provide the opportunity for future validation studies.

12.- **CIORDIA, M.**, & **GONZÁLEZ, A. J.** (2023). First report of *Neopestalotiopsis* sp. causing trunk disease on European chestnut in Spain. *Plant Disease 107*(10), 3307. DOI: 10.1094/PDIS-05-23-0987-PDN (OA) [CHF] [Ver online]

Abstract: In April 2021, a brownish-brown vascular lesion was observed in the stemwood of a five-year-old chestnut (*Castanea sativa Mill*.) rootstock when cutting for grafting in Villaviciosa (Asturias, Northern Spain). To identify the causal agent, a cross section of the steam was cut, surface-sterilized with 96% ethanol and dried, plated on potato dextrose agar (PDA) and incubated at 25C. Fungal colonies were consistently isolated and after 5 days developed abundant greyish-white mycelium. For molecular identification, the internal transcribed spacer (ITS) gene region of rDNA of the strain LPPAF-975 was amplified with the ITS1/ITS4 primers (White et al. 1990) using the TerraTM PCR Direct Polymerase Mix (Takara Bio Company, CA, USA). This sequence was deposited in GenBank (accession no: OR002144) and showed 99.80% identity over a 507 pb alignment with the *Neopestalotiopsis* isolate 328-16 (accession no: OK166668) isolated on blueberry in Serbia and *Nespestalotiopsis* australis strains LNZH0701 and LNZH0752 (accession nos: OM919511-12) both isolated on blueberry in China. To clarify the identification, beta-tubulin (tub2) and translation elongation factor alpha-1 (*tef1*-a) were amplified (Glass & Donaldson 1995, Walker et al. 2010, respectively). Beta-tubulin sequence (accession no: OR001747) was 99.52% of identity with sequences of several species of *Neopestalotiopsis*, and elongation factor (accession no: OR001748) was 99.57% identity with sequences previously deposited for *N. clavispora* (accession nos: OP684010-11, MZ097377-79). The phylogenetic tree was constructed with the three concatenated sequences using the Maximum Likelihood method based on the Tamura-Nei model (Tamura and Nei, 1993), and their topological robustness was evaluated by

bootstrap analysis based on 1,000 replicates using Mega 11 (Tamura et al, 2021). However, strain LPPAF-975 clustered with *N. javaensis*, *N. rosae* and *N. vacciniicola*, therefore the species remains unidentified. Pathogenicity tests were carried out on ten five-year-old chestnut plants on which a half 5-mm-diameter PDA plug from the edge of an actively growing colony of the fungus was inoculated by a cut in the bark of one to three branches per plant and then wrapped with Parafilm©. Five plants inoculated as above but without the fungus were used as controls. Plants were cultivated in pots provided with drip irrigation in a tunnel under natural conditions. The assay was conducted twice. External cankers around the inoculated area were observed a month after inoculation, while control plants did not show any lesions. The fungus was re-isolated on all the inoculated plants, but not from controls. As all the re-isolated strains had the same morphology, one of them was randomly selected and identified by sequencing, thus fulfilling Koch's postulates. Cross-sections of plants showed lesions similar to that initially found, with 100% of the plants damaged at the point of inoculation, and 80 and 65% 1 cm above and below, respectively. Pathogen was newly re-isolated and identified from one of these cross-sections. To the best of our knowledge, this is the first report worldwide of *Neopestalotiopsis sp*. causing disease on *Castanea sativa*. This pathogen could represent a threat to the maintenance of the biodiversity of traditional chestnut varieties, multiplied by grafting on rootstocks in nurseries, which would incur considerable economic losses.

13.- **DE LA ROZA-DELGADO, B.**, **MARTÍNEZ-FERNÁNDEZ, A.**, **MODROÑO, M.S.**, & **ARGAMENTERIA, A**. (2023). Sown meadows of *Lolium perenne* and *Trifolium repens* in Asturias. III. Ingestion and fill value in dairy cattle. *ITEA-Información Técnica Económica Agraria,119*(4), 357-369. DOI: 10.12706/itea.2023.009. (OA) [NPF] [Ver online]

Abstract: Digestibility, energy and nitrogen balances for eight years were determined in green forage of perennial ryegrass and white clover meadows, and in addition to first cut (in trench silo) and second cut (in big bales) silages. The temporary grassland was characteristic of the central coastal land of Asturias (Atlantic Area). The trials were carried out on individualized Frisian cows in metabolism stalls. Six production animals participated in each one with ad libitum forage. These animals were divided into three groups according to management: two of them with forage as the only diet (control group) and four supplemented with concentrate (three different concentrates) twice a day, across the four dosing scales throughout the years. The relative ingestibility, fill units and substitution rate were determined. The relative ingestibility (scale 0-1) of green forage was 0.87 in March and decreased to 0.71 in October ($p < 0.05$), while in terms of fill units/kg dry matter, they results were 1.01-1.24, respectively. For first and second cut silages, the relative ingestibilities were 0.70 and 0.82, respectively, and the milk fill units were 1.26 and 1.08 (not significant). The substitution rate was 0.37, without significant differences between concentrates. Results from this study suggest that the evaluated temporary grassland has a very good potential for voluntary forage intake in dairy cattle, with an adequate response to supplementation with concentrate.

14.- **DÍEZ, M.C.**, PRZYBORSKI, S., **DEL CERRO, A.**, ALONSO, M., IGLESIAS, T., **CARROCERA, S.**, **GARCÍA, M.A.**, FERNÁNDEZ, M., ALONSO, L. & **MUÑOZ, M**. (2023). Generation of a novel three-dimensional scaffold-based model of the bovine endometrium. *Veterinary Research Communications 47*(3), 1721-1733. DOI: 10.1007/s11259-023-10130-0 (OA) [GRA] [SA] [Ver online]

Abstract: Bovine in vitro endometrial models that resemble tissue function in vivo are needed to study infertility, long-term uterine alterations induced by pathogens and impact of endocrine disruptor chemicals on reproductive function and other reproductive system complications that cause high economic losses in livestock species. The present study aimed to generate an innovative, reproducible, and functional 3D scaffold-based model of the bovine endometrium structurally robust for long term-culture. We developed a multicellular model containing both endometrial epithelial and stromal cells. Epithelial cells organized to form a luminal-like epithelial layer on the surface of the scaffold. Stromal cells produced their own extracellular matrix forming a stable subepithelial compartment that physiologically resembles the normal endometrium. Both cell types released prostaglandin E2 and prostaglandin F2alpha following a treatment with oxytocin and arachidonic acid. Additionally signal pathways mediating oxytocin and arachidonic acid stimulation of prostaglandin synthesis were analyzed by real time PCR (RT-PCR). *Oxytocin receptor (OXTR), prostaglandin E2 receptor 2 (EP2*), *prostaglandin E2 receptor 4(EP4*), prostaglandin F receptor (PTGFR), *prostaglandin E synthase (PTGES),* PGF-synthase (PGFS) and prostaglandin-endoperoxide synthase 2 (*COX-2*) expression was detected in both control and treatment groups, however, only significant changes in abundance of *OXTR* mRNA transcripts were found. The results obtained by this study are a step forward in bovine in vitro culture technology. This 3D scaffold-based model provides a platform to study regulatory mechanisms involved in endometrial physiology and can set the basis for a broader tool for designing and testing novel therapeutic strategies for recurrent uterine pathologies.

15.- **ESPÍ, A., DEL CERRO, A.**, PEÓN, P., GONZÁLEZ, J.V., **SOMOANO, A.** (2023). Ticks and Tick-Borne zoonotic pathogens from wild birds in Northwestern coastal Spain. *Zoonotic Diseases, 3*(4), 316-333. DOI:10.3390/zoonoticdis3040026 (OA) [SA] [Ver online]

Abstract: Migratory and local birds may disperse ticks and their associated pathogens. The aim of this study was to provide information regarding ticks infesting birds in Asturias, a region that accounts for most of the Lyme disease hospitalizations in Spain. From September 2021 to April 2023, trained and experienced bird-banders collected ticks from birds in two estuary and four forest locations. A total of 1698 birds (52 species, 38 genera, and 26 families) were captured. A total of 51 ticks (28 larvae, 20 nymphs, and 3 females) were collected from 43 birds, belonging to three species: *Ixodes ricinus* (31), *Ixodes frontalis* (18), and *Haemaphysalis concinna* (1). The average prevalence of tick infestation was 2.5% and the average tick burden was 1.2 ticks per infested host. The bird species *Turdus merula*, *Parus major*, *Luscinia svecica*, and *Anthus pratensis* were among the most infested. *Anaplasma phagocytophilum* was detected in one *I. ricinus* nymph collected from *Phylloscopus collybita*. We have not detected *Borrelia burgdorferi* s.l., *Rickettsia* spp., *Coxiella burnetii*, or piroplasmids in any of the 51 analyzed ticks. These results suggest low infestation rate in migratory/estuary birds and a higher rate in forest/sedentary ones. Despite this, the detection of pathogens, although with low prevalence, can pose a risk to public health.

16.- **FORCADA, S.**, **MENÉNDEZ-MIRANDA, M.**, BOENTE, C., RODRÍGUEZ GALLEGO, J. L., COSTA-FERNÁNDEZ, J. M., **ROYO, L. J.**, & SOLDADO, A. (2023). Impact of potentially toxic compounds in cow milk: how industrial activities affect animal primary productions. *Foods, 12*(8), 1718. DOI: 10.3390/foods12081718 (OA) [NPF] [Ver online]

Abstract: Potentially toxic elements (PTEs) and polycyclic aromatic hydrocarbons

(PAHs) frequently coexist in soils near industrial areas and sometimes in environmental compartments directly linked to feed (forage) and food (milk) production. However, the distribution of these pollutants along the dairy farm production chain is unclear. Here, we analyzed soil, forage, and milk samples from 16 livestock farms in Spain: several PTEs and PAHs were quantified. Farms were compared in terms of whether they were close to (<5 km) or far away from (>5 km) industrial areas. The results showed that PTEs and PAHs were enriched in the soils and forages from farms close to industrial areas, but not in the milk. In the soil, the maximum concentrations of PTEs reached 141, 46.1, 3.67, 6.11, and 138 mg kg-1 for chromium, arsenic, cadmium, mercury, and lead, respectively, while fluoranthene (172.8 g kg-1) and benzo(b)fluoranthene (177.4 g kg-1) were the most abundant PAHs. Principal component analysis of the soil PTEs suggested common pollution sources for iron, arsenic, and lead. In the forage, the maximum contents of chromium, arsenic, cadmium, mercury, and lead were 32.8, 7.87, 1.31, 0.47, and 7.85 mg kg-1, respectively. The PAH found in the highest concentration in the feed forage was pyrene (120 g kg-1). In the milk, the maximum PTE levels were much lower than in the soil or the feed forages: 74.1, 16.1, 0.12, 0.28, and 2.7 g kg-1 for chromium, arsenic, cadmium, mercury, and lead, respectively. Neither of the two milk samples exceeded the 20 g kg-1 limit for lead set in EU 1881/2006. Pyrene was the most abundant PAH found in the milk (39.4 g kg-1), while high molecular weight PAHs were not detected. For PTEs, the results showed that soil-forage transfer factors were higher than forage-milk ratios. Our results suggest that soils and forages around farms near industries, as well as the milk produced from those farms, have generally low levels of PTE and PAH contaminants.

17.- **FORCADA, S.**, **MENÉNDEZ-MIRANDA. M.**, STEVENS, F., **ROYO, L. J.**, PIERNA, J. A. F., BAETEN, V., & SOLDADO, A. (2023). Industrial impact on sustainable dairy farms: Essential elements, hazardous metals and polycyclic aromatic hydrocarbons in forage and cow's milk. *Heliyon 9*(10), art. e20977. DOI: 10.1016/j.heliyon.2023.e20977. (OA) [NPF] [Ver online]

Abstract: Sustainable dairy farms are characterised by the self-production of forage for animal feed. These farms are sometimes located near industrial areas, entailing a risk of food chain contamination with hazardous metals and polycyclic aromatic hydrocarbons (PAHs). Accordingly, evaluating the impact of pollution on forage and milk is of great interest. In this study, the effects of industrial factors on sustainable forage from 43 dairy farms and possible correlations between inorganic elements and PAHs were studied. Spearman's correlation and principal component analysis (PCA) were performed for the forage and milk. Most of the inorganic elements in the forage were below the maximum residual limits for cadmium (Cd) and lead (Pb), established in EU 2013/1275 and EU 2019/1869, respectively. However, arsenic (As) and mercury (Hg) levels were above their respective limits in the forage (EU 2019/1869). No milk samples exceeded the maximum residual limits for Pb (EU 488/2014) or Cd (EU 1881/2006) in dairy products. Heavy-weight PAHs (HW-PAHs, four or more aromatic rings) were detected in forage but not in milk. In the forage samples, HW-PAHs were positively correlated with Zn and Cd. In addition, some hazardous metals (chromium (Cr), iron (Fe), As, Hg, and Pb) also were positively correlated with Zn and Cd. Interestingly, no correlations were found between forage pollutants and milk, suggesting that these pollutants have a low transfer rate to milk. The PCA results highlighted the predominant contribution of PAHs to the global variance in forage samples collected at different distances from industrial areas. In milk, the contributions of hazardous metals and PAHs were more balanced than in forages. Finally, when distances to potential pollution sources were included in the PCA of forage samples, a negative correlation was observed between the former and the concentrations of HW-PAHs, Cd, and Zn, suggesting that thermal power plants and steel factory emissions were the main sources of polluting forage in this area.

18.- GACEM, S., CASTELLO-RUIZ, M., **HIDALGO, C. O**., **TAMARGO, C**., SANTOLARIA, P., SOLER, C., YANIZ, J. & SILVESTRE, M. A. (2023). Bull sperm SWATH-MS-Based proteomics reveals link between high fertility and energy production, motility structures, and sperm–oocyte interaction. *Journal of Proteome Research 14*(9), 3607-3624. DOI: 10.1021/acs.jproteome.3c00461. [SRA] [Ver online]

Abstract: The prediction of male or semen fertility potential remains a persistent challenge that has yet to be fully resolved. This work analyzed several *in vitro* parameters and proteome of spermatozoa in bulls cataloged as high- (HF; $n = 5$) and low-field (LF; $n = 5$) fertility after more than a thousand artificial inseminations. Sperm motility was evaluated by computer-assisted sperm analysis. Sperm viability, mitochondrial membrane potential (MMP) and reactive oxygen species (mROS) of spermatozoa were assessed by flow cytometry. Proteome was evaluated by the SWATH-MS procedure. Spermatozoa of HF bulls showed significantly higher total motility than the LF group (41.4% vs 29.7%). Rates of healthy sperm (live, high MMP, and low mROS) for HF and LF bull groups were 49% and 43%, respectively ($p > 0.05$). Spermatozoa of HF bulls showed a higher presence of differentially abundant proteins (DAPs) related to both energy production (COX7C), mainly the OXPHOS pathway, and the development of structures linked with the motility process (TPPP2, SSMEM1, and SPAG16). Furthermore, we observed that equatorin (EQTN), together with other DAPs related to the interaction with the oocyte, was overrepresented in HF bull spermatozoa. The biological processes related to protein processing, catabolism, and protein folding were found to be overrepresented in LF bull sperm in which the HSP90AA1 chaperone was identified as the most DAP. Data are available via ProteomeXchange with identifier PXD042286.

19.- **GARCÍA-FERNÁNDEZ, B**., DOLCET-SANJUAN, R., MICHELETTI, D., **ANTÓN-DÍAZ, M. J.**, SOLSONA, C., **FERNÁNDEZ, M**., ABAD, X. & **DAPENA, E**. (2023). Susceptibility evaluation to Fire Blight and Genome-Wide associations within a collection of Asturian Apple Accessions. *Plants*,*12*(23), art. 4068. DOI: 10.3390/plants12234068 (OA) [CHF] [Ver online]

Abstract: Fire blight, caused by *Erwinia amylovora*, is one of the most devastating apple diseases. The selection of cultivars of low susceptibility and the study of the genetic mechanisms of the disease play important roles in fire blight management. The susceptibility level to fire blight was evaluated in 102 accessions originating from Asturias, a cider-producing region located in the north of Spain with a wide apple germplasm. Evaluations took place under quarantine conditions using artificial inoculations of grafted plants. The results revealed wide variation in susceptibility responses and low-susceptible cultivars were identified. In addition, 91 cultivars were genotyped using the Affymetrix Axiom (R) Apple 480 K SNP array to conduct genome-wide association studies (GWAS). A statistically significant signal was detected on chromosome 10 using the multi-locus mixed model (MLMM). Two genes were identified as major putative candidate genes: a TIR-NBS-LRR class disease protein and a protein containing a development and cell death (DCD) domain. The outcomes of this study provide a promising source of information, particularly in the context of cider apples, and set a starting point for future genetic and breeding approaches.

20.- GIMÉNEZ-GARCÍA, A., ALLEN-PERKINS, A., BARTOMEUS, I., BALBI, S.,

KNAPP, J. L., HEVIA, V., WOODCOCK, B. A., SMAGGHE, G., **MIÑARRO, M.**, EERAERTS, M., COLVILLE, J. F., HIPÓLITO, J., CAVIGLIASSO, P., NATES-PARRA, G., HERRERA, J. M., CUSSER, S., SIMMONS, B. I., WOLTERS, V., JHA, S., FREITAS, B. M., HORGAN, F. G., ARTZ, D. R., SIDHU, C. S., OTIENO, M., BOREUX, V., BIDDINGER, D. J., KLEIN, A.-M., JOSHI, N. K., STEWART, R. I. A., ALBRECHT, M., NICHOLSON, C. C., O'REILLY, A. D., CROWDER, D. W., BURNS, K. L. W., NABAES JODAR, D. N., GARIBALDI, L. A., SUTTER, L., DUPONT, Y. L., DALSGAARD, B., DA ENCARNAÇÃO COUTINHO, J. G., LÁZARO, A., ANDERSSON, G. K. S., RAINE, N. E., KRISHNAN, S., DAINESE, M., VAN DER WERF, W., SMITH, H. G., & MAGRACH, A. (2023) Pollination supply models from a local to global scale. *Web Ecology, 23*, 99-129. DOI: 10.5194/we-23-99-2023 (OA) [CHF] [Ver online]

Abstract: Ecological intensification has been embraced with great interest by the academic sector but is still rarely taken up by farmers because monitoring the state of different ecological functions is not straightforward. Modelling tools can represent a more accessible alternative of measuring ecological functions, which could help promote their use amongst farmers and other decision-makers. In the case of crop pollination, modelling has traditionally followed either a mechanistic or a data-driven approach. Mechanistic models simulate the habitat preferences and foraging behaviour of pollinators, while data-driven models associate georeferenced variables with real observations. Here, we test these two approaches to predict pollination supply and validate these predictions using data from a newly released global dataset on pollinator visitation rates to different crops. We use one of the most extensively used models for the mechanistic approach, while for the data-driven approach, we select from among a comprehensive set of state-of-the-art machine-learning models. Moreover, we explore a mixed approach, where data-derived inputs, rather than expert assessment, inform the mechanistic model. We find that, at a global scale, machine-learning models work best, offering a rank correlation coefficient between predictions and observations of pollinator visitation rates of 0.56. In turn, the mechanistic model works moderately well at a global scale for wild bees other than bumblebees. Biomes characterized by temperate or Mediterranean forests show a better agreement between mechanistic model predictions and observations, probably due to more comprehensive ecological knowledge and therefore better parameterization of input variables for these biomes. This study highlights the challenges of transferring input variables across multiple biomes, as expected given the different composition of species in different biomes. Our results provide clear guidance on which pollination supply models perform best at different spatial scales - the first step towards bridging the stakeholder-academia gap in modelling ecosystem service delivery under ecological intensification.

21.- **GIMENO, I.**, SALVETTI, P., **CARROCERA, S.**, GATIEN, J., GARCÍA, P., LÓPEZ, C., VALLEDOR, L. & **GÓMEZ, E.** (2023). Biomarker metabolite mating of viable frozen-thawed IVP bovine embryos with pregnancy-competent recipients leads to improved birth rates. *Journal of Dairy Science 106*(9), 6515-6538. DOI: 10.3168/jds.2022-23082 (OA) [GRA] [Ver online]

Abstract: Selection of competent recipients before embryo transfer (ET) is indispensable for improving pregnancy and birth rates in cattle. However, pregnancy prediction can fail when the competence of the embryo is ignored. We hypothesized that the pregnancy

potential of biomarkers could improve with information on embryonic competence. In vitro produced (IVP) embryos cultured singly for 24 h (from d 6 to 7) were transferred to d 7 synchronized recipients as fresh or after freezing and thawing. Recipient blood was collected on d 0 (estrus; n = 108) and d 7 (4-6 h before ET; n=107) and plasma was analyzed by nuclear magnetic resonance (1H+NMR). Spent embryo culture medium (CM) was collected and analyzed by UHPLC-MS/MS in a subset of n=70 samples. Concentrations of metabolites quantified in plasma (n = 35) were statistically analyzed as a function of pregnancy diagnosed on d 40, d 62 and birth. Univariate analysis with plasma metabolites consisted of a block study with controllable fixed factors (i.e., embryo cryopreservation, recipient breed, and day of blood collection; Wilcoxon test and t-test). Metabolite concentrations in recipients and embryos were independently analyzed by iterations that reclassified embryos or recipients using the support vector machine (SVM). Iterations identified some competent embryos, but mostly competent recipients that had a pregnancy incompetent partner embryo. Misclassified recipients that could be classified as competent were reanalyzed in a new iteration to improve the predictive model. After subsequent iterations, the predictive potential of recipient biomarkers was recalculated. On d 0, creatine, acetone and l-phenylalanine were the most relevant biomarkers at d 40, d 62, and birth, and on d 7, l-glutamine, l-lysine, and ornithine. Creatine was the most representative biomarker within blocks (n =20), with a uniform distribution over pregnancy endpoints and type of embryos. Biomarkers showed higher abundance on d 7 than d 0, were more predictive for d 40 and d 62 than at birth, and the pregnancy predictive ability was lower with frozen-thawed (F-T) embryos. Six metabolic pathways differed between d 40 pregnant recipients for fresh and F-T embryos. Within F-T embryos, more recipients were misclassified, probably due to pregnancy losses, but were accurately identified when combined with embryonic metabolite signals. After recalculation, 12 biomarkers increased ROC-AUC (>0.65) at birth, highlighting creatine (ROC-AUC = 0.851), and 5 new biomarkers were identified. Combining metabolic information of recipient and embryos improves the confidence and accuracy of single biomarkers.

22.- **GONZÁLEZ, A.J**. & **CIORDIA, M**. (2023). Shoot dieback in thornless blackberries in Nothern Spain caused by *Diaporthe rudis* and *Gnomoniopsis idaeicola*. *Horticulturae, 9*(9), 965. DOI:10.3390/horticulturae9090965 (OA) [CHF] [Ver online]

Abstract: A cane disease of a non-commercial thornless blackberry cultivar (genus *Rubus*, subgenus *Rubus* Watson) obtained in a breeding program was observed in May 2021 in northern Spain during a field evaluation. Symptoms of the disease appeared in spring and firstly consisted of dark-brown lesions in the petioles, tips, and intermediate zones of the canes, finally causing the leaves, canes, and lateral shoots to die. Two strains were recovered from infected canes and identified by morphological characteristics and multigene analysis as *Gnomoniopsis idaeicola* (LPPAF-977) and *Diaporthe rudis* (LPPAF-981). Pathogenicity tests showed that both fungi caused shoot dieback when artificially inoculated, reproducing the symptoms originally observed. Moreover, tissue necrosis was enhanced when *Diaporthe rudis* and *Gnomoniopsis idaeicola* were co-inoculated. This is the first report of *Diaporthe rudis* and *Gnomoniopsis idaeicola* causing a potentially serious disease to blackberries in Spain.

23.- **GONZÁLEZ-BLANCO, L., SIERRA, V., DIÑEIRO, Y.,** COTO-MONTES, A**.,** & **OLIVÁN, M**. (2023). Role of the endoplasmic reticulum in the search for early biomarkers of meat quality. *Meat Science 203,* art. 109224. DOI: 10.1016/j.meatsci.2023.109224 (OA) [SPA] [Ver online]

Abstract: Defects in meat quality such as dark, firm and dry (DFD) beef have been related to high levels of oxidative stress that produce cellular alterations that may affect to the process of meat quality acquisition. Despite the important role of endoplasmic reticulum (ER) in the cellular response to oxidative stress, its function in the muscle-to-meat conversion process has not yet been studied. In this study, differences in muscular antioxidant defense and the unfolded protein response (UPR) of the ER in CONTROL (normal pH24) and dark, firm, and dry (DFD, $\mathrm{pH24} \geq 6.2$) beef at 24h *post-mortem* were analyzed to understand the changes in the muscle-to-meat conversion process related to meat quality defects. DFD meat showed poor quality, lower antioxidant activity ($P<0.05$) and higher UPR activation ($P<0.05$), which indicates higher oxidative stress what could partly explain the occurrence of meat quality defects. Therefore, the biomarkers of these cellular processes (IRE1alpha, ATF6alpha, and p-eIF2alpha) are putative biomarkers of meat quality.

24.- GONZÁLEZ, L., PÉREZ, A., QUIROGA, A. C., **TAMARGO, C**., RAMOS, P., & BERMEJO, P. (2023). Effect of arachidonic acid on pre-and post-hatching in vitro bovine embryo development. *Reproduction, Fertility, and Development*, *35*(12), 614-621. DOI: 10.1071/RD23053. (OA) [SRA] [Ver online]

Abstract: Context. Arachidonic acid (AA) is the precursor of prostaglandins, which may play autocrine roles during early embryo development. Aims. To test the developmental effects of addition of AA to pre- and post-hatching culture media on in vitro-produced bovine embryos. Methods. Pre-hatching effects of AA were tested by culturing bovine zygotes in synthetic oviductal fluid (SOF) supplemented with 100 or 333 mu M AA. Post-hatching effects of AA were tested by culturing Day 7 blastocysts in N2B27 supplemented with 5, 10, 20 or 100 mu M AA up to Day 12. Key results. Pre-hatching development to blastocyst was completely abrogated at 333 mu M AA, whereas blastocyst rates and cell numbers were not altered at 100 mu M AA. Impaired post-hatching development was observed at 100 mu M AA, whereas no effect on survival rates was noted at 5, 10 and 20 mu M AA. However, a significant reduction in Day 12 embryo size was observed at 10 and 20 mu M AA. Hypoblast migration, epiblast survival and formation of embryonic-disc-like structures were unaffected at 5-10 mu M AA. AA exposure downregulated the genes *PTGIS*, PPARG, *LDHA* and *SCD* in Day 12 embryos. Conclusions. Pre-hatching embryos are mostly irresponsive to AA, whereas AA was observed to have negative effects during early post-hatching development. Implications. AA does not improve in vitro bovine embryo development and is not required up to early post-hatching stages.

25.- GU, S.H., **MIÑARRO, M**., FELIU, C., HUGOT, J-P., FORRESTER N.L., WEAVER, S.C., YANAGIHARA, R. (2023). Multiple lineages of hantaviruses harbored by the Iberian Mole (*Talpa occidentalis*) in Spain. *Viruses, 15*, art. 1313. DOI: 10.3390/v15061313 (OA) [CHF] [Ver online]

Abstract: The recent detection of both Nova virus (NVAV) and Bruges virus (BRGV) in European moles (*Talpa europaea*) in Belgium and Germany prompted a search for related hantaviruses in the Iberian mole (*Talpa occidentalis*). RNAlater-preserved lung tissue from 106 Iberian moles, collected during January 2011 to June 2014 in Asturias, Spain, were analyzed for hantavirus RNA by nested/hemi-nested RT-PCR. Pairwise alignment and comparison of partial L-segment sequences, detected in 11 Iberian moles from four parishes, indicated the circulation of genetically distinct hantaviruses. Phylogenetic analyses, using maximum-likelihood and Bayesian methods, demonstrated three distinct hantaviruses in Iberian moles: NVAV, BRGV, and a new hantavirus,

designated Asturias virus (ASTV). Of the cDNA from seven infected moles processed for next generation sequencing using Illumina HiSeq1500, one produced viable contigs, spanning the S, M and L segments of ASTV. The original view that each hantavirus species is harbored by a single small-mammal host species is now known to be invalid. Host-switching or cross-species transmission events, as well as reassortment, have shaped the complex evolutionary history and phylogeography of hantaviruses such that some hantavirus species are hosted by multiple reservoir species, and conversely, some host species harbor more than one hantavirus species.

26.- HERRERO, G., ACEVEDO, P., QUIRÓS, P., **PRIETO, J.M.**, ROMERO, B., AMADO, J., QUEIPO, M.A., GORTÁZAR, C.,& BALSEIRO, A. (2023). Tuberculosis epidemiology and spatial ecology at the cattle-wild boar interface in Northern Spain. *Transboundary and Emerging Diseases*, (2023), art. 2147191. DOI:10.1155/2023/2147191 (OA) [SA] [Ver online]

Abstract: Tuberculosis (TB) is a contagious chronic disease due to infection with *Mycobacterium tuberculosis* complex (MTC) bacteria. Monitoring of wildlife, especially potential reservoirs, is important for detecting changes in disease occurrence and assessing the impact of interventions. Here, we examined whether wild boar (*Sus scrofa*) may contribute to the re-emergence of TB in Asturias (10,604 km(2)), northern Spain. Although this province was declared free of TB in cattle in November 2021, MTC bacteria remain prevalent in several "hotspots," with the European badger (*Meles meles*) suggested as a TB potential wild reservoir. Drawing on data from the Spanish National Bovine Tuberculosis Eradication Program and the Government of the Principality of Asturias covering the period 2014-2020, we analyzed the prevalence of TB in cattle and wild boar in this region. In hotspots (592 km(2)), we also investigated the ranging behavior and habitat use of five cows that belonged to farms with a history of TB and six trapped sympatric wild boar. During the observation period, TB prevalence was 0.14% among cattle overall and 0.13-0.41% in hotspots, which was much lower than the prevalence in wild boar, which was 3.15% overall and 5.23-5.96% in hotspots. Infected cattle and infected wild boar in hotspots shared the same strains of M. bovis, and GPS tracking showed spatiotemporal overlap between the species, mainly around pastures during sunrise (06:00-07:00 h) and sunset (19:00-20:00 h). Our results suggest that in addition to cattle and badgers, wild boar possibly help maintain TB in northern Spain, increasing the host richness that influences TB transmission risk in the area, which should be taken into account in monitoring and eradication efforts.

27.- **JURADO, M., GARCÍA FERNÁNDEZ, C., CAMPA, A**., & **FERREIRA, J. J**. (2023). Genetic erosion within the Fabada dry bean market class revealed by high-throughput genotyping. *The Plant Genome*, *16*(4), art. e20379. DOI: 10.1002/tpg2.20379 (OA) [CHF] [Ver online]

Abstract: The Fabada market class within the dry beans has a well-differentiated seed phenotype with very large white seeds. This work investigated the genetic diversity maintained in the seed collections within this market class and possible genetic erosion over the last 30 years. A panel with 100 accessions was maintained in seed collections for 30 years, 57 accessions collected from farmers in 2021, six cultivars developed in SERIDA, and 16 reference cultivars were gathered and genotyped with 108,585 SNPs using the genotyping-by-sequencing method. Filtering based on genotypic and phenotypic data was carried out in a staggered way to investigate the genetic diversity among populations. The dendrogram generated from genotyping revealed 90 lines forming 16 groups with identical SNP profiles (redundant lines) from 159 lines classified

as market-class Fabada according to their passport data. Seed phenotyping indicated that 19 lines were mistakenly classified as Fabada (homonymies), which was confirmed in the dendrogram built without redundant lines. Moreover, this study provides evidence of genetic erosion between the population preserved for 30 years and the currently cultivated population. The conserved population contains 54.6% segregation sites and 41 different SNP profiles, whereas the cultivated population has 19.6% segregation sites and 26 SNP profiles. The loss of genetic variability cannot be attributed to the diffusion of modern cultivars, which increase genetic diversity (six new SNP profiles). The results allow for the more efficient preservation of plant genetic resources in genebanks, minimizing redundant accessions and incorporating new variations based on genotypic and phenotypic data.

28.- JUSTE, R., **BLANCO VÁZQUEZ, C.**, BARRAL, M., **PRIETO, J.M.**, VARELA, L., LESELLIER, S., DAVE, D., SEVILLA, I., MARTIN, A.B., ADRIAENSEN, H., HERRERO, G., GARRIDO, J., **CASÁIS, R.** & BALSEIRO, A. (2023) Efficacy of heat-inactivated *Mycobacterium bovis* vaccine delivered to European badgers (*Meles meles*) through edible bait. *Heliyon, 9*, art. 19349. DOI: 10.1016/j.heliyon.2023.e19349 (OA) [SA] [Ver online]

Abstract: Badgers (*Meles meles*) are a major tuberculosis (TB) reservoir in Europe, with the potential to transmit infection to cattle. Here we assessed whether a recently described oral tuberculosis vaccine based on heat-inactivated Mycobacterium bovis (HIMB), delivered as edible baits, can protect badgers from infection. Eight badgers were given individually five baits, each one consisting of a ball of peanut butter, natural peanut and oat flakes including a dose of the vaccine containing 5*107colony-forming units. In parallel, a control group of seven badgers did not receive the vaccine. One month and a half later a second dose of the vaccine was offered to the vaccinated group. Ninety-four days after the second dose, all badgers were challenged with *M. bovis* (103colony-forming units per animal) delivered endobronchially to the right middle lung lobe. Clinical, immunological, pathological and bacteriological variables were measured throughout the whole study to assess the efficacy of the vaccine. Two vaccinated animals showed high bacterial load of M. bovis and worsening of pathological lesions of TB. Conversely, the other six vaccinated animals showed slight improvement in bacterial load and pathology with respect to the control group. These results suggest that delivering the TB vaccine via food bait can partially protect wild badger populations, although vaccination can lead to either protection or tolerization, likely depending on the animal's immune status and general condition at the time of vaccination. Further optimization of the vaccination trial/strategy is needed to reduce the rate of tolerization, such as altering vaccine dose, number of doses, type of bait, use of adjuvants or route of administration.

29.- **LÓPEZ, R.**, SÁNCHEZ, O., PASCUAL, E., BORREL, Y. J., MARTÍNEZ, J. y ARIAS, A. (2023). *Plicopurpura patula* and *Monoplex parthenopeus* (Gastropoda: Caenogastropoda): New evidence of the tropicalization of the Bay of Biscay (NE Atlantic). *Journal of Sea Research, 194,* 1-11. DOI:10.1016/j.seares.2023.102412 (OA) [SA] [Ver online]

Abstract: Recent changes in climate and oceanographic conditions have been affecting the dynamics and composition of species communities worldwide, increasing the occurrence of tropical-subtropical species within a specific biogeographic area. This phenomenon defined as tropicalization is occurring in the Cantabrian Sea (Bay of Biscay), a transitional zone between two different regions of the North Atlantic. In this study we present new records of the tropical-subtropical gastropod species *Plicopurpura patula and Monoplex parthenopeus* in the Eu-ropean Atlantic, which constitute their northernmost distribution in the Atlantic Ocean to date. We provide a detailed characterization of both species, combining morphological and molecular features. Finally, supported by provided evidence and justification, we propose both species as two new potential bioindicators of the tropi-alization phenomenon in the North Atlantic waters.

30.- MARTÍNEZ-HERNÁNDEZ, J., COLÍN-NAVARRO, V., **ROSA-GARCÍA, R**., ALBARRÁN-PORTILLO, B., VALDÉS-PIÑA, M. G., ARRIAGA-JORDÁN, C. M., & FLORES, J. G. E. (2023). Livestock in the sustainability of changing livelihoods in high mountain communities in a Natural Protected Area in Central Mexico. *Agro Productividad, 8* . DOI: 10.32854/agrop.v16i8.2565. (OA) [NPF] [Ver online]

Abstract: Livelihoods of communities within the high mountain Natural Protected Area for Flora and Fauna 'Nevado de Toluca' (NPAFFNT) in the central highlands of Mexico have changed due to statuary limitations established in management program respect to agro-silvopastoral activities. In these communities, the small-scale farmers who still rely on livestock, mainly sheep production. There is no knowledge on the sustainability, strengths and weaknesses of these systems and the influence of seasons in this area of marked bioclimatic seasonality. Methodology: The IDEA method to assess sustainability was applied in a small village within NPAFFNT in the dry as in the rainy season. Results: Sustainability is limited by the economic scale while their main strengths were in the agroecological scale in both seasons. Indicators show deficiencies in farmer education, lack of technical assistance, poor local infrastructures, lack of access to institutional support programmes, and the revision of norms for the natural protected area. Conclusions and implications: Results are useful for improving the sustainability of these systems that may improve que quality of life of high-mountain communities and provide ecosystemic services relevant for society. Diversification of income sources are of needed given the limitations that the new statute creates on livestock and agricultural activities. It is advisable to increase the educational level of the inhabitants and farmers receive specialized technical training in agricultural and sustainable livestock production.

31.- MENGA, V., FARES, C., **CAMPA, A**., **FERREIRA, J.J**., BITOCCHI, E., PAPA, R., & BELEGGIA, R. (2023). Variability of nutritional, antioxidant, and textural traits of a collection of snap beans of different colors. *Horticulturae 9*(3), 311. DOI: 10.3390/horticulturae9030311 (OA) [CHF] [Ver online]

Abstract: A set of 54 snap bean lines grown under organic farming was characterized for different traits: pod color, pod cross-section shape, pod section width (PSW), protein content (PC), and sugar content (S). After cooking, the lines were analyzed for firmness and color parameters (CIE-L*, a*, b*). The snap bean lines were grouped based on pod cross-section shape and pod color into eight groups, and significant differences were observed among lines and groups for all the traits investigated. In particular, the yellow pods were harder than the other snap beans and less sweet due to the negative correlation between firmness and sugar content. Fourteen selected lines with contrasting firmness and belonging to different color groups were investigated for their phenolic composition and antioxidant activity (TEAC) before and after domestic cooking. A general decrease was observed only for vanillic acid, quercetin, and apigenin-8-C-glucoside. Quercetin-3-O-rutinoside and kaempferol-3-O-rutinoside were detected in all samples with the highest values in the SBP042 line in both raw and cooked samples. Antioxidant activity decreased with cooking (average of 39%), but purple line SBP053 showed the lowest and no significant loss (3.1%). The results reported in this study could be useful to design specific varieties for different markets and purposes.

32.- **MIÑARRO, M.**, GARCÍA, D. & **ROSA GARCÍA, R.** (2023). Pollination of exotic fruit crops depends more on extant pollinators and landscape structure than on local management of domestic bees. *Agriculture, Ecosystems and Environment, 347*, art.108387 DOI: 10.1016/j.agee.2023.108387 (OA) [CHF] [NPF] [Ver online]

Abstract: Agricultural globalization has driven the expansion of exotic crops into new agricultural areas. Pollinator -dependent exotic crops not only have to face the abiotic constraints of the new cultivation regions but also deal with local pollinator assemblages, which may or may not fulfill pollination requirements. Here, we studied how three entomophilous exotic crops (kiwifruit, northern highbush blueberry and rabbiteye blueberry) adapt to a common pollination environment in new cultivation areas in Spain. For this, we assessed the pollination limitation of those crops, the contribution of insect assemblages to the pollination service, and the effect of landscape structure and the management of domestic pollinators on these assemblages. The three exotic crops showed large and diverse pollinator assemblages but differed in the assemblage composition and in the identity of the main pollinator species. Honeybee clearly dominated kiwifruit assemblages, representing almost 70 % of visits to flowers. Bumblebees and honeybee fairly equally dominated floral visits in highbush blueberry, and bumblebees accounted for more than 90% of visits in rabbiteye blueberry. Floral morphology partially explained interspecific differences in pollinator assemblages and led to the distinct contributions of the different insects to the different crops. Kiwifruit (but not blueberry) crops experienced pollination limitation that led to, on average, a 7.2 % reduction in fruit weight. This pollination limitation decreased when honeybee abundance rose. In all three crops, the local pollinator assemblages mostly depended on the landscape structure around orchards but were only affected by the management of domestic pollinators in rabbiteye blueberry crops. Our results highlight the importance of understanding the interspecific differences in the pollination ecology of new exotic crops before designing general management recommendations, and also question the use of managed pollinators before making an assessment of the contribution of extant insects to the pollination service.

33.- **NAVARRO, A., MUÑOZ, M.**, **IGLESIAS, N.**, **BLANCO- VÁZQUEZ**, C., BALSEIRO, A., MILHANO SANTOS, F., CIORDIA, S., CORRALES, F.J., **IGLESIAS, T.**, & **CASAIS, R.** (2023^{a}). Proteomic serum profiling of Holstein Friesian cows with different pathological forms of bovine paratuberculosis reveals changes in the acute-phase response and lipid metabolism. *Journal of Proteome Research*. DOI: 10.1021/acs.jproteome.3c00244. (OA) [SA] [GRA] [Ver online]

Abstract: The lack of sensitive diagnostic methods to detect *Mycobacterium avium* subsp. *paratuberculosis* (Map) subclinical infections has hindered the control of paratuberculosis (PTB). The serum proteomic profiles of naturally infected cows presenting focal and diffuse pathological forms of PTB and negative controls (*n* = 4 per group) were analyzed using TMT-6plex quantitative proteomics. Focal and diffuse are the most frequent pathological forms in subclinical and clinical stages of PTB, respectively.

One (focal versus (vs.) control), eight (diffuse vs. control), and four (focal vs. diffuse) differentially abundant (DA) proteins (q-value < 0.05) were identified. Ingenuity pathway analysis of the DA proteins revealed changes in the acute-phase response and lipid metabolism. Six candidate biomarkers were selected for further validation by specific ELISA using serum from animals with focal, multifocal, and diffuse PTB-associated lesions *(n* = 108) and controls (*n* = 56). Overall, the trends of the serum expression levels of the selected proteins were consistent with the proteomic results. Alpha-1-acid glycoprotein (ORM1)-based ELISA, insulin-like growth factor-binding protein 2 (IGFBP2)-based ELISA, and the anti-Map ELISA had the best diagnostic performance for detection of animals with focal, multifocal, and diffuse lesions, respectively. Our findings identify potential biomarkers that improve diagnostic sensitivity of PTB and help to elucidate the mechanisms involved in PTB pathogenesis.

34.- OJEDA-MARÍN, C., GUTIÉRREZ, J. P., FORMOSO-RAFFERTY, N., **GOYACHE, F**., & CERVANTES, I. (2023ª- 2024^{p}). Differential patterns in runs of homozygosity in two mice lines under divergent selection for environmental variability for birth weight. *Journal of Animal Breeding and Genetics 141*(2*).* DOI: 10.1111/jbg.12835. (OA) [GRA] [Ver online]

Abstract: Runs of homozygosity (ROH) are defined as long continuous homozygous stretches in the genome which are assumed to originate from a common ancestor. It has been demonstrated that divergent selection for variability in mice is possible and that low variability in birth weight is associated with robustness. To analyse ROH patterns and ROH-based genomic inbreeding, two mouse lines that were divergently selected for birth weight variability for 26 generations were used, with: 752 individuals for the high variability line (H-Line), 766 individuals for the low variability line (L-Line) and 74 individuals as a reference population. Individuals were genotyped using the high density Affymetrix Mouse Diversity Genotyping Array. ROH were identified using both the sliding windows (SW) and the consecutive runs (CR) methods. Inbreeding coefficients were calculated based on pedigree (FPED) information, on ROH identified using the SW method (FROHSW) and on ROH identified using the CR method (FROHCR). Differences in genomic inbreeding were not consistent across generations and these parameters did not show clear differences between lines. Correlations between FPED and FROH were high, particularly for FROHSW . Moreover, correlations between FROHSW and FPED were even higher when ROH were identified with no restrictions in the number of heterozygotes per ROH. The comparison of FROH estimates between either of the selected lines were based on significant differences at the chromosome level, mainly in chromosomes 3, 4, 6, 8, 11, 15 and 19. ROH-based inbreeding estimates that were computed using longer homozygous segments had a higher relationship with FPED . Differences in robustness between lines were not attributable to a higher homozygosis in the L-Line, but maybe to the different distribution of ROH at the chromosome level between lines. The analysis identified a set of genomic regions for future research to establish the genomic basis of robustness.

35.-PÉREZ. T., ROMERO, A., PIRHADI, N., COYA, R., FERNÁNDEZ, M.ª.P., **MÁRQUEZ, I.**, GARCÍA, L. & BORRELL, Y. (2023). Insights on the evolutionary history and genetic patterns of *Octopus vulgaris* Cuvier, 1797 in the Northeastern Atlantic using mitochondrial DNA. *Animals, 13*(17), art. 2798. DOI: 10.3390/ani13172708 (OA) [SA] [Ver online]

Abstract: *Octopus vulgaris* is one of the most harvested octopus species in the world. In the Iberian Peninsula, there are several small-scale fisheries that have a long-term

tradition of harvesting octopus. The Asturias fleet (in Northern Spain) has an internationally recognized MSC label for its exploitation. Of concern, genetic assessments of exploited stocks are currently scarce, which could prevent the implementation of adequate managing strategies. We use two mitochondrial regions (cytochrome oxidase subunit 1 and control region) to analyze the genetic status and evolutionary events that conditioned octopus populations' characteristics in the Northeastern Atlantic. A total of 90 individuals were sampled from three different localities in the Iberian Peninsula as well as a location in Macaronesia. Temporal genetic analyses on Asturias and Algarve populations were also performed. Results indicated the absence of fine spatial genetic structuring but showed the Canary Islands (in Macaronesia) as the most distinct population. Our analyses detected two distinct clades, already described in the literature, but, for the first time, we confirmed the presence of the alpha-southern haplogroup in the Northern Iberian Peninsula. This result indicates a more continuous cline for the distribution of these two haplogroups than previously reported. Temporal changes in the distribution of both haplogroups in contact zones were also detected.

36.- PIRHADI, N., PARRONDO, M., ROMERO-BASCONES, A., THOPPIL, R., MARTÍNEZ, J. L., FERNÁNDEZ-RUEDA, M. P., **MÁRQUEZ, I**., GARCÍA-FLÓREZ, L., DOPICO, E., PÉREZ, T., & BORRELL, Y. J. (2023). Genetic monitoring on the world's first MSC eco-labeled common octopus (*O. vulgaris*) fishery in western Asturias, Spain. *Scientific Reports*, *13*(1), art. 2730. DOI: 10.1038/s41598-023-29463-6 (OA) [SA] [Ver online]

Abstract: *Octopus vulgaris* (Cuvier, 1797) is a cephalopod species with great economic value. In western Asturias (northwest of Spain), O. vulgaris artisanal fisheries are relatively well monitored and conditionally eco-labeled by the Marine Stewardship Council (MSC). Despite this, the Asturian octopus stocks have not been genetically assessed so far. In order to improve the current fishery plan and contrast the octopus eco-label validity in Asturias, 539 individuals from five regions of the O. vulgaris geographic distribution, including temporal samplings in Asturias, were collected and genotyped at thirteen microsatellite loci. All the samples under analysis were in agreement with Hardy-Weinberg expectations. Spatial levels of genetic differentiation were estimated using F-statistics, multidimensional scaling, and Bayesian analyses. Results suggested that the O. vulgaris consists of at least four genetically different stocks coming from two ancestral lineages. In addition, temporal analyses showed stability in terms of genetic variation and high NE (>50) for several generations in different localities within Asturias, pointing out to indeed sustainable fishery exploitation levels. Even though, the current Asturias fishery plan shows no significant genetic damages to the stocks, the regional-specific management plans need systematic genetic monitoring schemes as part of an efficient and preventive regional fishery regulation strategy.

37.- **RODRÍGUEZ MADRERA, R., CAMPA, A**., & **FERREIRA, J. J.** (2023^{p}-2024^{a}). Modulation of the nutritional and functional values of common bean by farming system: organic vs conventional. *Frontiers in Sustainable Food Systems, 7*, art. 1282427. DOI: 10.3389/fsufs.2023.1282427 (OA) [TA] [CHF] [Ver online]

Abstract: Common bean (*Phaseolus vulgaris* L.) is a crop of high nutritional interest which is widespread throughout the world as a result of which it is considered as a potential crop for ensuring future food security. The effect of organic and conventional farming systems on the nutritional and functional composition of two cultivars of market class fabada (a landrace and a modern elite cultivar) is described. The findings of our study have shown that the common beans from organic farming have a higher content of protein, phenolic compounds, and relevant microelements such as iron and zinc than the seeds from conventional farming trial. However, in conventional crops a higher content of polyunsaturated fatty acids and lower concentrations of phytic acid and raffinose oligosaccharides were also detected. In contrast, no significant differences were detected in the proximate composition of the two genotypes tested, nor in their antioxidant activity or levels of phenolic compounds. The elite cultivar maintains the nutritional characteristics of the landrace from which it derives, suggesting the genomic regions modified in the improvement do not play an important role in their genetic control.

38.- **RODRÍGUEZ MADRERA, R**., & **PANDO, R**. (2023). The phenolic composition, antioxidant activity and microflora of wild elderberry in Asturias (Northern Spain): An untapped resource of great interest. *Antioxidants*, *12*(11), art.1986. DOI: 10.3390/antiox12111986 (OA) [TA] [Ver online]

Abstract: The objective of this study is the characterization of the phenolic profile and antioxidant activity of elderberries (*Sambucus nigra* L.) from a collection of 79 wild specimens in northern Spain to assess variations in the species at the local level and evaluate its interest as a source of biocompounds. Also, a first study was carried out on the microflora present in this fruit, providing information relevant to its commercial exploitation. Moreover, the phenolic composition, antioxidant capacity and microbial composition in overripe fruits were determined, seeking a better use for this currently wasted resource. A wide variability in levels of phenolics was detected. Elderberries showed high antioxidant activity related to a high cyanidin derivative content, making them of interest to industry. Microflorae were present in very variable concentration ranges, so their levels should be monitored in those applications that require strict control. Overripe fruits are of interest as a source of anthocyanidins, since their concentration and antioxidant capacity remain after the optimal ripening period, promoting sustainability and a better use of natural resources. The database generated is of particular interest for further breeding trials based on the phenolic profile and antioxidant activity of the samples.

39.- RUIZ-RUBIO, S., ORTIZ-LEAL, I., TORRES, M. V., **SOMOANO, A.**, & SÁNCHEZ-QUINTEIRO, P. (2023). Do fossorial water voles have a functional vomeronasal organ? A histological and immunohistochemical study. *The Anatomical Record*. *2023* DOI:10.1002/ar.25374 (OA) [SA] [Ver online]

Abstract: The fossorial water vole, *Arvicola scherman*, is an herbivorous rodent that causes significant agricultural damages. The application of cairomones and alarm pheromones emerges as a promising sustainable method to improve its integrated management. These chemical signals would induce stress responses that could interfere with the species regular reproductive cycles and induce aversive reactions, steering them away from farmlands and meadows. However, there is a paucity of information regarding the water vole vomeronasal system, both in its morphological foundations and its functionality, making it imperative to understand the same for the application of chemical communication in pest control. This study fills the existing gaps in knowledge through a morphological and immunohistochemical analysis of the fossorial water vole vomeronasal organ. The study is primarily microscopic, employing two approaches: histological, using serial sections stained with various dyes (hematoxylin-eosin, Periodic acid-Schiff, Alcian blue, Nissl), and immunohistochemical, applying various markers that provide morphofunctional and structural information. These procedures have confirmed the presence of a functional vomeronasal system in fossorial water voles, characterized by a high degree of differentiation and a significant expression of cellular markers indicative of active chemical communication in this species.

40.-SALAS-HUETOS, A., RIBAS-MAYNOU, J., MATEO-OTERO, Y., **TAMARGO, C**., LLAVANERA, M., & YESTE, M. (2023). Expression of miR-138 in cryopreserved bovine sperm is related to their fertility potential. *Journal of Animal Science and Biotechnology*, *14*(1), art. 129. DOI: 10.1186/s40104-023-00909-1 (OA) [SRA] [Ver online]

Abstract: Background: MicroRNAs (miRNAs) are small, single-stranded, non-coding RNA molecules of 22-24 nucleotides that regulate gene expression. In the last decade, miRNAs have been described in sperm of several mammals, including cattle. It is known that miRNAs can act as key gene regulators of early embryogenesis in mice and humans; however, little is known about the content, expression, and function of sperm-borne miRNAs in early bovine embryo. In this study, total sperm RNA was isolated from 29 cryopreserved sperm samples (each coming from a separate bull) using a RNeasy kit and treatment with DNase I. RNA concentration and purity were determined through an Epoch spectrophotometer and an Agilent Bioanalyzer. The expression of 10 candidate miRNAs in bovine sperm (bta-miR-10a, bta-miR-10b, bta-miR-138, bta-miR-146b, bta-miR-19b, bta-miR-26a, bta-miR-34a, bta-miR-449a, bta-miR-495 and bta-miR-7), previously identified in testis and/or epididymis, was evaluated with RT-qPCR. The cel-miR-39-3p was used as a spike-in exogenous control. Nonparametric Mann-Whitney tests were run to evaluate which miRNAs were differentially expressed between bulls with high fertility [HF; non-return rates (NRR) ranging from 39.5 to 43.5] and those with subfertility (SF; NRR ranging from 33.3 to 39.3). Several sperm functionality parameters (e.g., viability, membrane stability or oxygen consumption, among others) were measured by multiplexing flow cytometry and oxygen sensing technologies. Results: RNA concentration and purity (260/280 nm ratio) (mean & PLUSMN; SD) from the 29 samples were 99.3 & PLUSMN; 84.6 ng/& mu;L and 1.97 & PLUSMN; 0.72, respectively. Bioanalyzer results confirmed the lack of RNA from somatic cells. In terms of the presence or absence of miRNAs, and after applying the Livak method, 8 out of 10 miRNAs (bta-miR-10b, -138, -146b, -19b, -26a, -449a, -495, -7) were consistently detected in bovine sperm, whereas the other two (bta-miR-10a, and -34a) were absent. Interestingly, the relative expression of one miRNA (bta-miR-138) in sperm was significantly lower in the SF than in the HF group ($P = 0.038$). In addition to being associated to fertility potential, the presence of this miRNA was found to be negatively correlated with sperm oxygen consumption. The expression of three other miRNAs (bta-miR-19b, bta-miR-26a and bta-miR-7) was also correlated with sperm function variables. Conclusions: In conclusion, although functional validation studies are required to confirm these results, this study suggests that sperm bta-miR-138 is involved in fertilization events and beyond, and supports its use as a fertility biomarker in cattle.

41.- SALMAN, A., FERNÁNDEZ-ALEGRE, E., FRANCISCO-VÁZQUEZ, R., GÓMEZ, R., FERNÁNDEZ-FERNÁNDEZ, A., AREÁN-DABLANCA, H.,

DOMÍNGUEZ, J.C., GONZÁLEZ, J.R., **CAAMAÑO, J.N**.& MARTÍNEZ-PASTOR, F. (2023). Extension of the equilibration period up to 24 h maintains the post-thawing quality of Holstein bull semen frozen with OPTIXcell. *Animal Reproduction Science*, *250*, art.107209. DOI: 10.1016/j.anireprosci.2023.107209. (OA) [SRA] [Ver online]

Abstract: Semen cryopreservation in bovine livestock is well established, but logistics often require deviations from standard protocols. Extending the equilibration time to the following day is convenient in many situations. To improve our knowledge of the effects of this modification, we studied the post-thawing and post-incubation (4h, 38°C) sperm quality after freezing with 4 or 24-h extension in the OPTIXcell extender by using an ample panel of analyses: CASA for motility; flow cytometry for viability, physiology, oxidative stress, and chromatin parameters (DNA fragmentation, chromatin compaction, and thiol groups status); and spectrometry for malondialdehyde production. Semen was obtained from 12 Holstein bulls. The 24-h equilibration time showed few significant effects, with only a tiny decrease in progressive motility and a positive impact on chromatin structure. The incubation removed some of these effects, with the pattern for chromatin compaction remaining the same. No detrimental oxidative stress or increase in apoptotic or capacitation markers was detected. Additionally, the individual bull interacted with the effects of the incubation and the equilibration, especially regarding the chromatin status. Whereas this interaction did not critically affect sperm quality, it could be relevant in practice. Bull fertility as non-return rates (NRR56) was associated with some sperm parameters (especially with an improved chromatin structure) but not in the 4-h post-thawing analysis. Our study supports that extending the equilibration time by at least 24-h is feasible for bull semen freezing with the OPTIXcell extender.

42.- SALMAN, A., FERNÁNDEZ-ALEGRE, E., FRANCISCO-VÁZQUEZ, R., DOMÍNGUEZ, J. C., ÁLVAREZ-RODRÍGUEZ, M., **CAAMAÑO, J. N.**, MARTÍNEZ-PASTOR, F., GÓMEZ-MARTÍN, R., FERNÁNDEZ-FERNÁNDEZ, A. & AREÁN-DABLANCA, H. (2023). Pre-freezing selection of Holstein bull semen with the BoviPure colloid as double-or single-layer centrifugation improves the post-thawing quality. *Animal Reproduction Science*,*258*, art. 107344. DOI: 10.1016/j.anireprosci.2023.107344 [SRA] [Ver online]

Abstract: Artificial insemination (AI) is critical for breeding in the dairy industry. High-merit bulls can present low freezability, hampering genetic dissemination. Thawed semen can be improved using density gradient centrifugation (DGC) with colloids, but little information deals with the pre-freezing application. Thus, the BoviPure colloid (optimized for bull spermatozoa) was tested for pre-freezing application as the usual double-layer (DLC) versus single-layer (SLC, quick and economical). Semen from twelve Holstein-Friesian bulls was extended with OPTIXcell extender, frozen (Control), or processed by SLC or DLC and frozen. Sperm were assessed pre-freezing for motility and viability and post-thawing (directly and after 4h 38°C) for apoptosis, capacitation status, acrosomal damage, mitochondrial activity, cytoplasmic and mitochondrial reactive oxygen species (ROS), and chromatin status (SCSA for DNA fragmentation and chromatin compaction and monobromobimane, mBBr, for disulfide bridges evaluation). The DGC improved parameters post-thawing (e.g., 57.5%±10.1 motility vs. control 53.3%±11.2) at the cost of sperm loss (sperm recovery of DGC 14.4%±2.5 and SLC 17.4%±2.5). DNA fragmentation (%DFI) decreased (0.21%±0.53 vs. control 1.30%±0.10), and SLC reduced chromatin compaction. A clustering procedure separated lesser (LF) and greater freezability (GF) bulls. LF samples were especially benefited by DGC, with SLC providing better post-thawing results for this group. In conclusion, pre-freezing DGC improved sperm parameters post-thawing, potentially improving the cryopreservation of low-freezability semen from high-merit bulls. SLC, quicker and economical, would be preferable since it showed similar or higher performance than DLC.

43.-**TAMARGO, C**., SALMAN, A., **CAAMAÑO, J. N**., MARTÍNEZ-PASTOR, F., **FERNÁNDEZ, Á.**, MUIÑO, R., CARBAJO, M.T. & **HIDALGO, C. O**. (2023). Characterization of the germplasm bank for the spanish autochthonous bull breed "Asturiana de la Montaña" *Animals*,*13*(8), art. 1402. DOI: 10.3390/ani13081402 (OA) [SRA] [Ver online]

Abstract: Semen cryobanks are critical for preserving autochthonous and rare breeds. Since sperm cryopreservation has been optimized for commercial breeds, non-commercial ones (often endangered) must be characterized to ensure the germplasm's viability. This study reports an investigation of the "Asturiana de la Montaña" breed (AM), a valuable Spanish autochthonous cattle breed adapted to the mountainous Atlantic environment. The survey included cryopreserved semen doses from 40 bulls stored at the Principado de Asturias Germplasm Bank. Data were obtained from the routine fresh semen analysis, CASA (motility), and flow cytometry analyses of fresh and post-thawing semen, and the 56-day non-return-rate (NRR) in heifers and cows (all results as 1st and 3rd quartiles). Fresh samples (artificial vagina) were within the normal range for cattle (4-6 mL, 5-10 * 109/mL; mass motility 5). Post-thawing results showed motility below typical for commercial breeds (total motility 26-43%, progressive 14-28%), with higher values for viability (47-62%). Insemination results showed a good performance for this breed (NRR: 47-56%; higher for heifers). Sperm volume increased with age, with little or no effects on sperm quality. Few associations were found between post-thawing quality or freezability and NRR, LIN being the variable more strongly associated (positively). The AM semen bank shows a good prospect for preserving and disseminating the genetics of this breed. This survey indicates that dedicated research is needed to adapt freezing protocols to this breed, optimizing post-thawing results.

44.-VERDÚ, J., CORTEZ, V., **ROSA GARCÍA, R.**, ORTIZ, A., **GARCÍA-PRIETO, U.,** LUMARET, J.P., GARCÍA-ROMERO, C., & SÁNCHEZ-PIÑERO, F. (2023). Nontoxic effects of thymol, carvacrol, cinnamaldehyde, and garlic oil on dung beetles: A potential alternative to ecotoxic anthelmintics. *PLoS ONE 18*(12), art. e0295753. DOI: 10.1371/journal.pone.0295753 (OA) [NPF] [SPA] [Ver online]

Abstract: The sustainability of the traditional extensive livestock sector will only be possible if healthy dung-decomposing insect communities are preserved. However, many current pharmaceutical anthelmintics are harmful to dung beetles, their presence can have a negative impact on biological systems. Phytochemical anthelmintics are an alternative to ecotoxic synthetic pharmaceutical anthelmintics, although ecotoxicological tests of their possible indirect effects on dung beetles are required to demonstrate their viability. In this study, the potential ecotoxicity of thymol, carvacrol, cinnamaldehyde and garlic oil (diallyl disulfide and diallyl trisulfide) were tested for the first time. Inhibition of antennal response was measured as a relevant parameter by obtaining relevant toxicity thresholds derived from concentration–response curves, such as the IC50. All phytochemical compounds tested were demonstrated to be suitable alternative candidates to the highly ecotoxic compound ivermectin, considering their non-toxicity to nontarget organisms. Residues of the phytochemical antiparasitics found in cattle droppings were extremely low, even undetectable in the case of diallyl disulfide

and diallyl trisulfide. Furthermore, our results showed that none of the phytochemical compounds have ecotoxic effects, even at extremely high concentrations, including those almost 1000 times higher than what is most likely to be found in dung susceptible to ingestion by dung beetles in the field. We can conclude that the four selected phytochemical compounds meet the requirements to be considered reliable alternatives to ecotoxic veterinary medicinal products, such as ivermectin.

ÍNDICES

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- PANDO BEDRIÑANA, Rosa (TA): *(38)*
- PRIETO MARTÍN, José Miguel (SA): *(26) (28)*
- RODRÍGUEZ MADRERA, Roberto (TA): *(11), (37), (38)*
- ROSA GARCÍA, Rocío (NPF): *(30), (32), (44)*
- ROYO MARTÍN, Luís José (NPF): *(1), (8), (16), (17)*
- SIERRA SÁNCHEZ, Verónica: *(23)*
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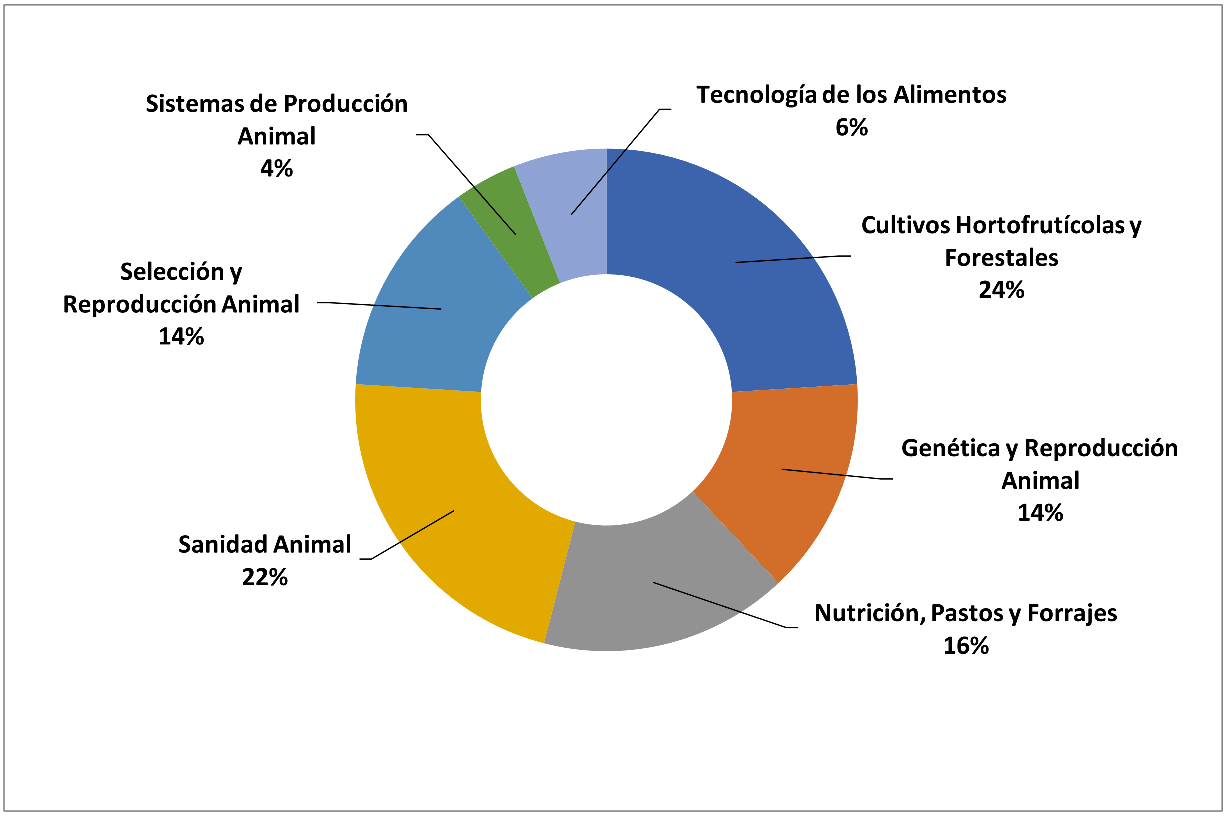
	Nº artículos	CUARTIL	FI 2022	Referencia	Área
Agriculture, Ecosystems and Environment	1	Q1	6,6	(32)	CHF NPF
Anatomical Record	1	Q2	2	(39)	SA
Agro Productividad	1	-	-	(30)	NPF
Animal Reproduction Science	2	Q2	2,2	(41)(42)	SRA (2)
Animals	3	Q1	3	(4)(35)(43)	GRA SA SRA
Antioxidants	1	Q1	7	(38)	TA
BMC Genomics	1	Q1	4,4	(7)	SA
BMC Plant Biology	1	Q1	5,3	(11)	CHF TA
Foods	2	Q1	5,2	(1) (16)	NPF (2)
Forest	1	Q1	2,9	(6)	CHF
Frontiers in Sustainable Food Systems	1	Q2	4,7	(37)	TA CHF
Frontiers in Veterinary Science	1	Q1	3,2	(2),	SA
Genetics Selection Evolution	1	Q1	4,1	(3)	GRA
Heliyon	3	Q2	4	(8)(17)(28)	NPF(2) SA
Horticulturae	2	Q1	3,1	(22)(31)	CHF(2)
ITEA: InfomaciónTécnica Económica Agraria	1	Q4	0,4	(13)	NPF
Journal of Animal Breeding and Genetics	2	Q1	2,6	(5)(34)	GRA(2)
Journal of Animal Science and Biotechnology	1	Q1	7	(40)	SRA
Journal of Dairy Science	1	Q1	3,5	(21)	GRA
Journal of Proteome Research	2	Q1	4,4	(18)(33)	SRA SA GRA
Journal of Sea Research	1	Q2	2	(29)	SA

Meat Science	1	Q1	7,1	(23)	SPA
Nature Communications	1	Q1	16,6	(9)	CHF
Plant Disease	1	Q1	4,5	(12)	CHF
Plants	1	Q1	4,5	(19)	CHF
PLoS ONE	1	Q2	3,7	(44)	NPF SPA
Reproduction, Fertility and Development	1	Q4	1,9	(24)	SRA
Scientific Reports	1	Q2	4,6	(36)	SA
The Plant Genome	1	Q1	4,2	(27)	CHF
Theriogenology	1	Q3	2,8	(10)	SRA
Transboundary and Emerging Diseases	1	Q1	4,3	(26)	SA
Veterinary Research Communications	1	Q2	2,2	(14)	GRA SA
Viruses	1	Q2	4,7	(25)	CHF
Web Ecology	1	Q3	2,2	(20)	CHF
Zoonotic Diseases (2021_)	1	-	-	(15)	SA

ARTÍCULOS POR ÁREAS DE INVESTIGACIÓN DEL SERIDA

- ÁREA DE CULTIVOS HORTOFRUTÍCOLAS Y FORESTALES: *(3), (9), (11), (12), (19), (20), (22), (25), (27), (31), (32), (37).*
- ÁREA DE GENÉTICA Y REPRODUCCIÓN ANIMAL: *(4), (5), (6), (14), (21), (33), (34).*
- ÁREA DE NUTRICIÓN, PASTOS Y FORRAJES: *(1), (8), (13), (16), (17), (30), (32), (44).*
- ÁREA DE SANIDAD ANIMAL: *(2), (7), (14), (15), (26), (28), (29), (33), (35), (36), (39).*
- ÁREA DE SELECCIÓN Y REPRODUCCIÓN ANIMAL: *(10), (18), (24), (40), (41), (42), (43).*
- ÁREA DE SISTEMAS DE PRODUCCIÓN ANIMAL: *(23), (44).*
- ÁREA DE TECNOLOGÍA DE LOS ALIMENTOS*: (11), (37), (38)*

Figura 1: Artículos por áreas de investigación del SERIDA



Fuente: Elaboración propia

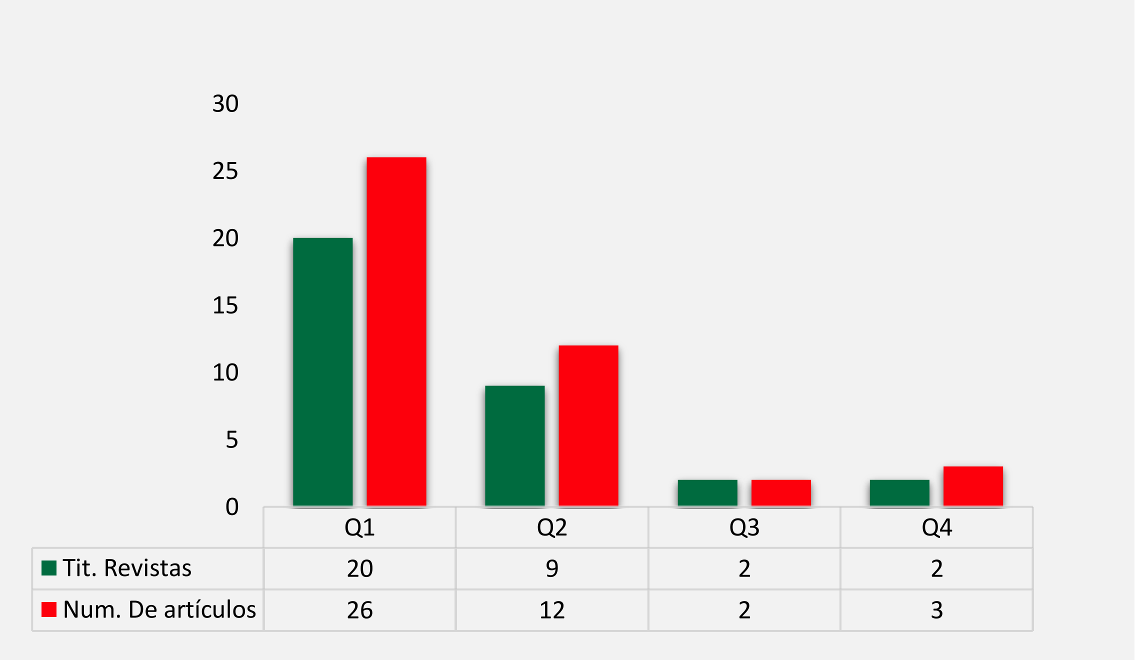
Figura 2: Número de artículos realizados en colaboración con otros países

Fuente: Web of Science

Figura 3: Número de artículos por título de revista

Fuente: Web of Science

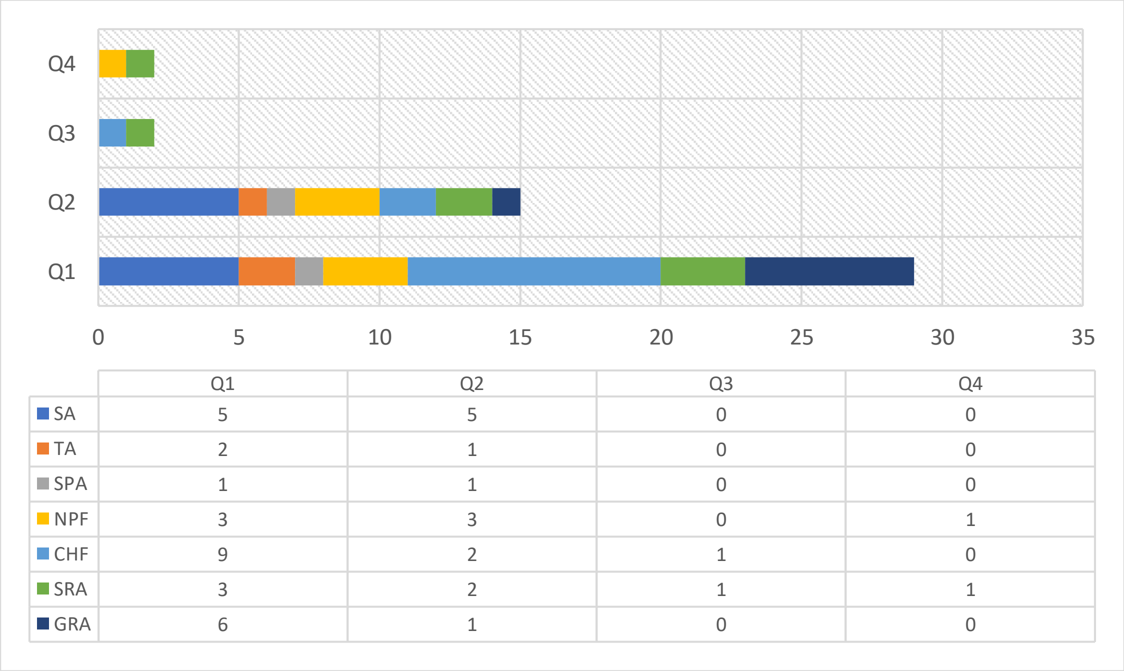
Figura 4: Cuartiles de las revistas en las que aparecen publicados los artículos del SERIDA



	Q1	Q2	Q3	Q4
🟩 Tit. Revistas	20	9	2	2
🟥 Num. De artículos	26	12	2	3

Fuente: Elaboración propia basada en JCR 2022

Figura 5: Cuartiles de las revistas en las que aparecen publicados los artículos del SERIDA según las distintas áreas de investigación



	Q1	Q2	Q3	Q4
🟦 SA	5	5	0	0
🟧 TA	2	1	0	0
⬜ SPA	1	1	0	0
🟨 NPF	3	3	0	1
🟦 CHF	9	2	1	0
🟩 SRA	3	2	1	1
🟦 GRA	6	1	0	0

Fuente: Elaboración propia basada en JCR 2022