Associazione Scientifica Italiana di Coniglicoltura

ATTI - *PROCEEDINGS*

GIORNATE DI CONIGLICOLTURA ASIC 2015

Forlì, 15-16 aprile 2015

Salone internazionale avicolo e cunicolo - edizione 2015

In collaborazione con

ALMA MATER STUDIORUM
UNIVERSITÀ DI BOLOGNA
DIPARTIMENTO DI SCIENZE E TECNOLOGIE AGRO-ALIMENTARI

In collaboration with

Sponsors

PRESENTAZIONE DELLE GIORNATE DI CONIGLICOLTURA ASIC 2015

Cari amici e soci ASIC,

siamo giunti alla sesta edizione delle Giornate di Coniglicoltura ASIC che, anche quest'anno, sono tenute in concomitanza con la Fiera Internazionale Avicola e Cunicola di Forlì. Il successo delle precedenti edizioni ci ha incoraggiato a continuare nell'impegnativo compito di consolidare il valore scientifico e dare maggiore visibilità internazionale a questa manifestazione.

L'evento, frutto della collaborazione tra ASIC e Ente Fiera di Forlì e patrocinato dall'ASPA e dal Dipartimento di Scienze e Tecnologie Agro-Alimentari dell'Università di Bologna, è stato realizzato anche grazie all'impegno dei membri del Comitato organizzatore e del Comitato scientifico. Un particolare apprezzamento va alla Fiera di Forlì che ci accoglie nelle proprie strutture e contribuisce alle spese per la traduzione e per l'ospitalità dei relatori invitati. Porgiamo un sentito ringraziamento ai partecipanti al convegno e alle aziende che hanno voluto sostenere economicamente l'iniziativa: il loro contributo permette di coprire i costi congressuali e aiuterà la nostra Associazione nella promozione di nuove iniziative formative e scientifiche.

Un caloroso benvenuto è infine rivolto a tutti i ricercatori che, credendo nell'importanza di questo congresso, hanno inviato i risultati delle loro ultime ricerche. Particolarmente apprezzata è la presenza di ricercatori e tecnici stranieri provenienti per questa edizione da Ungheria e Francia.

Le Giornate di Coniglicoltura ASIC 2015 si sviluppano in due giornate. La prima giornata si apre con una tavola rotonda incentrata sulle sfide che la coniglicoltura italiana sta affrontando in questi ultimi anni in relazione a sicurezza alimentare e uso dei farmici, sistemi di stabulazione e benessere animale, qualità dei prodotti e commercializzazione. Nello specifico, il Dott. Guido Grilli (Università degli Studi di Milano) presenta i risultati del Piano Antibiotici nazionale; il Dott. Silvio Borrello (Direzione Generale della Sanità Animale e dei Farmaci Veterinari) illustra la posizione del Ministero della Salute sulle recenti Linee di indirizzo inerenti il benessere dei conigli in allevamento, mentre la Dott.ssa Rossella Pedicone (UNAITALIA) e il Dott. Antonio Lavazza (Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna) ne presentano gli aspetti tecnici; infine, il Prof. Massimiliano Petracci (Università di Bologna) pone l'accento sulle caratteristiche nutrizionali delle carni cunicole, mentre il Dott. Marco Guerrieri (COOP Italia) presenta la posizione della carne di coniglio presso la grande distribuzione.

La seconda giornata prevede due relazioni ad invito: la prima "Sistemi di allevamento alternativi per le coniglie fattrici" è presentata dal Prof. Zsolt Szendrő (Università di Kaposvar, Ungheria) e introdotta dal Prof. Alessandro Dal Bosco (Università di Perugia) con breve presentazione su un progetto europeo sul benessere dei conigli (ANIHWA, Development and assessment of alternative animal-friendly housing systems for rabbit does with kits and growing rabbits); nella seconda relazione il Dott. Fabrizio Agnoletti (Istituto Zooprofilattico Sperimentale delle Venezie) presenterà "Nuove acquisizioni per la classificazione della virulenza e della capacità diffusiva di S. aureus nel coniglio". Il programma proseguirà con comunicazioni scientifiche nelle aree di Genetica, Sistemi produttivi, e Patologia. In concomitanza con le pause è prevista la sessione Poster.

Oltre alle relazioni ad invito, gli Atti del convegno racchiudono l'insieme dei contributi scientifici presentati in forma orale e di poster, 16 lavori complessivamente. Tutti i contributi sono stati sottoposti a referaggio da parte del Comitato Scientifico.

Fiera di Forlì, 15-16 Aprile 2015

Prof. Claudio Cavani
Presidente Comitato organizzatore

Prof. Antonio Camarda
Presidente ASIC

PRESENTATION OF GIORNATE DI CONIGLICOLTURA ASIC 2015

Dear Colleagues,

It is a pleasure to present you to the 6th edition of the Giornate di Coniglicoltura ASIC held in Forlì on April 15-16, 2015.

The meeting has been organized during the International Fair specialized in Poultry and Rabbit, in collaboration with Ente Fiera di Forlì.

The meeting took place on two days and included a round table and invited lectures addressed to both productive and scientific world as well as scientific sessions with communications and poster presentation.

The round table was focused on the challenge that Italian rabbit production chain must afford in the next years in the field of animal welfare, food quality and safety, and rabbit meat marketing.

Moreover two invited lectures were presented and focused on key-topics for the rabbit sector: “Alternative housing systems for rabbit does” (Prof. Zsolt Szendrő, University of Kaposvár, Hungary) and “New knowledge about the classification of virulence and diffusive ability of *S. aureus* in rabbit” (Dr. Fabrizio Agnoletti, Istituto Zooprofilattico Sperimentale delle Venezie).

Finally, scientific communications and posters were presented in the areas of Reproduction and Genetics, Nutrition and Physiology, Pathology, Welfare and Management and Meat Quality.

The abstracts of invited paper and scientific communications are presented here.

Fiera di Forlì, April 15-16, 2015

Prof. Claudio Cavani
President of Organizing Committee

Prof. Antonio Camarda
President of ASIC

GIORNATE DI CONIGLICOLTURA ASIC 2015

Associazione Scientifica Italiana di Coniglicoltura

Fiera di Forlì, 15-16 aprile 2015

ENTI ORGANIZZATORI – *PARTNERS*

Associazione Scientifica Italiana di Coniglicoltura (ASIC)

In collaborazione con – In collaboration with:

FIERA DI FORLÌ – Fieravicola Edizione 2015

COMITATO ORGANIZZATORE – *ORGANIZING COMMITTEE*

Prof. Claudio Cavani (Presidente), Dipartimento di Scienze e Tecnologie Agro-Alimentari, Università di Bologna.

Dott. Luca Bano, Istituto Zooprofilattico delle Venezie, Sez. di Treviso.

Prof. Antonio Camarda, Dipartimento di Medicina Veterinaria, Università di Bari.

Prof. Alessandro Dal Bosco, Dipartimento di Biologia Applicata. Sez. di Scienze Zootecniche, Università di Perugia.

Dott. Francesco Danese, Veronesi Verona spa.

Dott. Andrea Frabetti, Martini spa.

Dott.ssa Rossella Pedicone, Unaitalia.

Prof. Massimiliano Petracci, Dipartimento di Scienze e Tecnologie Agro-Alimentari, Università di Bologna.

Dott. Ivan Toschi, Dipartimento di Scienze Agrarie e Ambientali, Università di Milano.

Dott.ssa Angela Trocino, Dipartimento di Biomedicina Comparata e Alimentazione, Università di Padova.

COMITATO SCIENTIFICO – *SCIENTIFIC COMMITTEE*

Prof. Antonio Camarda (Presidente), Dipartimento di Sanità e Benessere degli Animali, Università di Bari.

Dott. Fabrizio Agnoletti, Istituto Zooprofilattico delle Venezie, Sez. di Treviso.

Dott. Guido Grilli, Dipartimento di Patologia Animale, Igiene e Sanità Pubblica Veterinaria, Università di Milano.

Dott. Antonio Lavazza, Istituto Zooprofilattico della Lombardia ed Emilia Romagna, Brescia.

Prof. Gerolamo Xiccato, Dipartimento di Agronomia Animali Alimenti Risorse Naturali e Ambiente, Università di Padova.

SEGRETERIA SCIENTIFICA – *SCIENTIFIC SECRETARY*

Prof. Massimiliano Petracci

Dipartimento di Scienze e Tecnologie Agro-Alimentari, Università di Bologna

Tel. +39 0547 338128 (Fax +39 0547 382348)

Dott.ssa Angela Trocino

Dipartimento di Biomedicina Comparata e Alimentazione, Università di Padova

Email: segreteria@asic-wrsa.it

PROGRAMMA

MERCOLEDÌ 15 APRILE 2015 (Sala Europa)

14:30-15:00 Saluto ai convenuti e Apertura dei lavori

Guido Sassi, Presidente della Fiera di Forlì

Antonio Camarda, Presidente dell'Associazione Scientifica Italiana di Coniglicoltura (ASIC)

Gerolamo Xiccato, Presidente WRSA

Claudio Cavani, Presidente del Comitato Organizzatore del Convegno

15:00-17:00 Tavola Rotonda: "Nuove sfide per il settore cunicolo: un'opportunità di promozione del consumo" *Moderatori: Prof .Gerolamo Xiccato, Prof. Antonio Camarda*

Interventi programmati:

- *Piano Antibiotici nazionale, presentazione dei risultati* (Dott. Guido Grilli, Università degli Studi di Milano).
- *Linee di indirizzo inerenti il benessere dei conigli in allevamento, la posizione del Ministero della Salute* (Dott. Silvio Borrello, Direzione Generale della Sanità Animale e dei Farmaci Veterinari
- *Linee di indirizzo inerenti il benessere dei conigli in allevamento, gli aspetti tecnici* (Dott.ssa Rossella Pedicone, UNAITALIA; Dott. Antonio Lavazza, Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna)
- *La qualità nutrizionale delle carni cunicole* (Prof. Massimiliano Petracci, Università di Bologna)
- *La carne di coniglio presso la grande distribuzione* (Dott. Marco Guerrieri, COOP Italia)

17:00-17:30: Coffee break offerto da una italia

A seguire: Assemblea dei soci ASIC

19:30 Incontro conviviale offerto da Elanco

GIOVEDÌ 16 APRILE 2015 (Sala Borsa)

9:00-9:15 Saluto ai convenuti e Apertura dei lavori

9:15-9:30 *ANIHWA, Development and assessment of alternative animal-friendly housing systems for rabbit does with kits and growing rabbits*, Prof. Alessandro Dal Bosco (Università di Perugia, Italia)

9:30-10:00 *Alternative housing systems for rabbit does*, Prof. Zsolt Szendrő (Università di Kaposvár, Ungheria)

10:00-10:30 *Nuove acquisizioni per la classificazione della virulenza e della capacità diffusiva di S. aureus nel coniglio*, Dott. Fabrizio Agnoletti (Istituto Zooprofilattico Sperimentale delle Venezie)

Sessione Poster

10:30-11:00

Dynamics of the serological response of commercial rabbits to Borghi and SG33 vaccine strains
Bano L., Marcon B., Drigo I., Tonon E., Fracas V., Mazzolini E., Dorigo F., Agnoletti F.

Antimicrobial susceptibility of clinical isolates of Escherichia coli in rabbit industrial herds in 2010-2014
Berto G., Mazzolini E., Bano L., Brunetta R., Gagliazzo L., Agnoletti F.

Environmental impact of rabbit production through Life Cycle Assessment
Cesari V., Negretti R., Zucali M., Bava L., Toschi I.

Susceptibility testing in Escherichia coli strains using the disk pre-diffusion, the disk diffusion tests and the minimum inhibitory concentration
Cocchi M., Deotto S., Di Sopra G., Di Giusto T.

Survey on biosecurity measures and farmers' awareness about antimicrobial resistance in industrial rabbit farms of the Veneto region
Di Martino G., Brunetta R., Gagliazzo L., Buniolo F., Brichese M., Agnoletti F., Bonfanti L.

A prototype of colony cage with removable walls for improving the welfare of rabbit does: preliminary behavioural results
Martino M., Mattioli S., Ruggeri S., Cambiotti V., Moscati L., Castellini C., Dal Bosco A.

Sessione Comunicazioni scientifiche (Sala Borsa)

Genetica e sistemi produttivi
Coordinatore: Alessandro Dal Bosco
11:00-12:00
A candidate gene strategy identified markers associated with growth rate in a meat rabbit line
Fontanesi L., Utzeri V.J., Scotti E., Fornasini D., S. Dall'Olio, Frabetti A.

The effect of fattening rabbit housing systems in mobile arks: relation between in vivo oxidative status and meat quality
Mattioli S., Martino M. , Ruggeri S., Moscati L., Dal Bosco A., Castellini C.

Effect of enrichment on carcass and meat quality in growing rabbits reared in collective pens
Trocino A., Maccarana L., Birolo M., Tazzoli M., Zuffellato A., Xiccato G.

Effect of feeding programme on performance and health of growing rabbits
Birolo M., Tazzoli M., Trocino A., Maccarana L., Xiccato G.

Patologia

Coordinatore: Antonio Camarda

12:00-13:00

Using DNA microarray technique in determining the virulence gene profile of rabbit Escherichia coli strains isolated from colibacillosis outbreaks in Northen Italy

Badagliacca P., Tonelli A., Drigo I., Pompilii C., Di Provvido A., Agnoletti F.

On short term the reduction of antimicrobial consumption in fattening rabbits seems not correlated with decreasing antimicrobial resistance

Berto G., Mazzolini E., Bano L., Brunetta R., Guolo A., Puiatti C., Agnoletti F.

Antimicrobial consumption in industrial rabbit farming, Italy, 2010 to 2013

Brunetta R., Mazzolini E., Bano L., Berto G., Bonfanti L., Di Martino G., Brichese M., De Rui S., Camerotto P., Brino A., Agnoletti F.

Concurrent infection of rabbit (*Oryctolagus cuniculus*) and Italian hare (*Lepus corsicanus*) by Rabbit Haemorrhagic Disease type 2 virus

Camarda A., Pugliese N., Cavadini P., Circella E., Capucci L., Caroli A., Legretto M., Mallia E., Lavazza A.

13:00-13:20 Sessione poster e Cerimonia di chiusura

13:20-14:30 Pranzo offerto da una italia

GIORNATE DI CONIGLICOLTURA ASIC 2015

Associazione Scientifica Italiana di Coniglicoltura

Fiera di Forlì, April 15-16th 2015

PROGRAMME

WEDNESDAY APRIL 15TH, 2015 (location Sala Europa)

14:30-15:00 Welcome and Opening session

Guido Sassi, President of Fiera Forlì

Antonio Camarda, President of Italian Association of Rabbit Science (ASIC)

Gerolamo Xiccato, President of World Rabbit Science Association (WRSA)

Claudio Cavani, President of Organizing Committee Giornate ASIC

15:00-17:00 Round table: "New challenges for the rabbit production system: an opportunity to promote rabbit meat consumption" - *Chairpersons: Prof .Gerolamo Xiccato, Prof. Antonio Camarda*

Scheduled interventions

- *National antibiotic plan, presentation of results* (Dr. Guido Grilli, University of Milan).
- *Guidelines on welfare of farmed rabbits, the position of the Health Ministry* (Dr. Silvio Borrello, Direzione Generale della Sanità Animale e dei Farmaci Veterinari)
- *Guidelines on welfare of farmed rabbits, technical issues* (Dr. Rossella Pedicone, UNAITALIA; Dr. Antonio Lavazza, Istituto Zooprofilattico Sperimentale della Lombardia e dell'Emilia Romagna)
- *Nutritional quality of rabbit meat* (Prof. Massimiliano Petracci, University of Bologna)
- *Rabbit meat and large scale distribution* (Dr. Marco Guerrieri, COOP Italia)

17:00-17:30: Coffee break offered by una italia

17:30-18:30 Plenary assembly of ASIC members

19:30 Social dinner offered by Elanco

THURSDAY APRIL 16TH 2015 (Sala Borsa)

9:00-9:15 Welcome and Opening session

9:15-9:30 *ANIHWA project - Development and assessment of alternative animal-friendly housing systems for rabbit does with kits and growing rabbits*, Prof. Alessandro Dal Bosco (University of Perugia, Italy)

9:30-10:00 *Alternative housing systems for rabbit does*, Prof. Zsolt Szendrő (University of Kaposvár, Hungary)

10:00-10:30 *New knowledge about the classification of virulence and diffusive ability of S. aureus in rabbits*, Dr. Fabrizio Agnoletti (Istituto Zooprofilattico Sperimentale delle Venezie).

Poster Session

10:30-11:00

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Oral communications

Genetics and production systems

11:00-12:15

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12:00-12:15

Effect of feeding programme on performance and health of growing rabbits

Birolo M., Tazzoli M., Trocino A., Maccarana L., Xiccato G.

Pathology

12:15-13:15

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Camarda A., Pugliese N., Cavadini P., Circella E., Capucci L., Caroli A., Legretto M., Mallia E., Lavazza A.

13:15-13:30 Poster session and Closing session

13:30-14:30 Lunch offered by

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Alternative housing systems for breeding does

Szendrő Zs., Matics Zs., Gerencsér Zs.

Kaposvár University, 40, Guba S. str, Kaposvár, H-7400, Hungary

Corresponding Author: Prof. Zsolt Szendrő, Kaposvár University, 40, Guba S. str, Kaposvár, H-7400, Hungary - Email: szendro.zsolt@ke.hu

The main results and observations on group and individually housed rabbit does were reviewed by Szendrő and McNitt (2012), however in the recent years several new data were published in this field. This gives a new opportunity to summarize the current knowledge about alternative housing systems for breeding does.

In Switzerland rabbit does are generally housed in group systems. The recent Belgian and Dutch housing systems will be converted step by step into group housing system. According to several experimental results currently there are no researcher who is able to offer a solution to eliminate aggression, stress and injures which are common among rabbit does in group housing systems. Relating to individual housing of rabbit does some authors are drawn to the opinion that these cages are small, the moving possibility and the social contact are limited. Positive results were published when platforms were inserted into the cages and the possibility for moving increased, the does and their kits could jump up and down. Using footrest the incidence of sore hock declined. Several environmental enrichments also can be used which increase the wellbeing of rabbit does. These enriched cages (equipped with platform, footrest, gnawing sticks, etc.) are fully in line with the animal welfare requirements.

Increasing the size of cages and enriching them increase the production cost and the meat will be more expensive. This is particularly true for the group housing systems. Developing housing systems we have to take into consideration this aspect as well, because the rabbit production in the EU could decline dramatically.

New insight from laboratory to classify the affecting and diffusing ability of *Staphylococcus aureus* in rabbit

Agnoletti F.

Istituto Zooprofilattico Sperimentale delle Venezie, Diagnostic and Microbiology Laboratory, Treviso, Italy

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Staphylococcus aureus (*S. aureus*) is a Gram-positive microorganism commonly colonising the skin of people and several animal species, but also capable to affect different organs and cause diseases from mild to quite severe thanks to a complex set of virulence factors. *S. aureus* is one of the most important causes of rabbits breeder stock replacement, due to pododermatitis, mastitis and fibrinous purulent metritis, whereas nests can be devastated by necrotic dermatitis and septicaemia. Occasionally rabbits of any age can be affected by subcutaneous abscesses, arthritis, rhinitis and fibrinous purulent pneumonia.

As it is for several rabbit diseases *S. aureus* outbreaks are often trigged by environmental (e.g. traumatizing cage's metal floors) or managerial factors (e.g. use or rabbit semen for artificial insemination contaminated by *S. aureus*) as well, but here the microorganism features play a pivotal role in the severity of the clinical picture and in the intra-herd prevalence. Moreover *S. aureus* is often resistant to several antimicrobials and recently the clone ST398 of *S. aureus* methicillin resistant (MRSA), which is considered one of the most relevant multi-drug resistant organisms (MDRO), was reported in rabbit meat industrial herds.

Several international initiatives have been implemented in Europe to fight the burden of antimicrobial resistance (AMR) and prompt for a strong reduction of antimicrobial consumption in food producing animals and for a specific usage ban for animal therapy of certain antimicrobials, for instance 3^{d} and 4th generation cephalosporins and macrolides, because they are of critical choice for humans therapy. The above mentioned scenario suggests that therapy is not the appropriate tool to control or reduce staphylococcosis prevalence in rabbit herds and that other prophylactic measures must be adopted.

If we consider the pyramidal organization of the rabbit meat production system a proper policy to tackle staphylococcosis would keep the grandparents/parents selection herds free from most virulent *S. aureus* clones and rabbit breeding flocks for meat production protected by vaccines or autovaccines.

Both eradication or vaccination strategies should be adopted and both require high discriminatory laboratory tools to distinguish clones of different virulence and diffusive ability within the wide *S. aureus* population. The latter ability is of great relevance as all rabbit farmers in each production cycle may experience sporadic and sometimes quite severe *S. aureus* infection at single rabbit level, however staphylococcosis turns into a farm problem when high disease prevalence and incidence occur. Regardless culling of affected animals, the diffusive ability of certain clones results in high staphylococcosis incidence and its persistence within flock.

S. aureus biotyping and phage typing were initially used to characterize aggressive clones. The phenotype biotype CV-type C, phage type 3A/3C/55/7 was recognized as the clone causing chronic

staphylococcosis and because of male and female breeder trade also more diffuse in Europe. Due to low intra-laboratories repeatability, being time and resources consuming, notwithstanding difficulties to standardize the method, phage typing was early dismissed by laboratories. More recently molecular methods have replaced phenotype characterisation and PCR detection of *bbp, selm* genes and the flank sequence is now used to screen high virulent *S. aureus* (HV-SA) isolates.
However this approach, although able to detect more aggressive clones, was set up in North-European countries and overall answer a "yes or no" question: whether the strain is an HV-SA.
We aimed to go further and verify whether the high virulence (i.e. capability to produce severe lesions) is also associated with high within flock diffusibility and whether clones reported as circulating in North Europe are also circulating in Italian herds. We developed a study aimed to verify the correlation between a) clinical data collected in a wide and representative set of rabbit meat herds and b) the molecular features of *S. aureus* strains collected in same herds. Forty herds were investigated during the study and over 2400 rabbits were clinically examined. Overall 850 *S . aureus* strains were collected and 400 of them further genotyped by *mecA, mecC, selm, bbp, cna, fnbB* and flank sequence PCR detection and by PFGE, MLST and *spa* typing.
The statistical analysis allowed to identify certain pathogenic and diffusive clusters (*spa* CC 645 and *spa* CC 024) and other not pathogenic (*spa* CC 1190 and *spa* CC094). In this study, among all methods used to characterise *S. aureus* rabbit virulent clones, *spa* typing displayed the highest discriminatory power. *Spa* typing is based on the sequencing of the X region of the *S. aureus* protein A coding gene. This method has an high intra and inter laboratories repeatability, the output is provided in highly portable data (a sequence) and a web based software is available to cluster bacterial strains according to *spa* homology, thus providing a valuable tool for *S. aureus* epityping.

A candidate gene strategy identified markers associated with growth rate in a meat rabbit line

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The identification of DNA markers associated with growth rate or related measures could open new possibilities to improve selection responses in rabbit breeding programs. In this study we applied a candidate gene approach to identify markers associated with finishing weight in a commercial meat rabbit line. The selected candidates were three genes that encode for key components of the somatotropic axis (insulin like growth factor 2, *IGF2*; growth hormone, *GH1*; and growth hormone receptor, *GHR*) that, in other species have been already shown to affect several production traits. The first step was the identification of polymorphisms. This was done by resequencing coding and non coding regions of the mentioned genes using amplified products obtained designing primer pair in the corresponding regions annotated on the OryCun2.0 genome version. In particular, all exons and parts of non coding regions (*GH1* and *GHR*) or only a few coding regions (*IGF2*) were sequenced in a panel of 10-12 rabbits of different breeds and lines. Alignment of the obtained sequences produced 20 single nucleotide polymorphisms and one indel. Then, a polymorphism for each gene was selected to genotype by PCR-RFLP 250-450 rabbits of a commercial meat line for which weight at 70 days was available. Association study between these mutations and finishing weight in the genotyped rabbits was carried out using the procedure MIXED of SAS, with a model that included the buck as a random effect and the fixed effects of sex, year, litter, and genotype. All genotyped polymorphisms were associated ($P<0.05$) with the investigated parameter. These results confirm that a candidate gene approach could successfully identify markers associated with production traits in rabbits opening new possibilities to design rabbit breeding programs including DNA marker information.

Key words: SNP, Polymorphisms, Growth rate, Association study.

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The effect of fattening rabbit housing systems in mobile arks: relation between *in vivo* oxidative status and meat quality

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The aim of this trial was to study the effect of an alternative housing system on the oxidative status and meat quality of fattening rabbits. From May to June 2015, 30 rabbits of 35 days of age were reared in mobile ark placed outdoor on alfalfa grass and weekly moved for 42 days. To assess the health status of animals, blood samples were collected at begin of the trial and at the slaughter on rabbits housed outdoors and in conventional cages (C). Meat quality parameters were also evaluated. Concerning the *in vivo* oxidative status, ark-reared rabbits showed higher TBARS (thiobarbituric reactive substances) values than C ones probably for the higher motory activity given by the larger living area; also the plasma α-tocopherol content was higher (4.75 *vs* 3.70 nmol/mL) most likely for grass consumption. Accordingly, carcasses obtained from ark-reared rabbits showed lower perirenal and perivisceral fat. Even the lipid content of LL (*Longissimus lumborum*) muscle was lower (1.08 *vs* 0.50%; $P<0.05$) in ark group. There were no significant differences in the LL muscle pH and colour, whereas water holding capacity and cooking loss were higher in ark than C group. Given the higher intake of bioactive compounds through the grass, the meat antioxidant content was higher in ark-reared rabbits (7.4 *vs* 6.8 ng/g of retinol, 719.2 *vs* 683.3 ng/g of α-tocopherol, respectively). The fatty acid profile of ark rabbits reflected the higher consumption of essential fatty acids by grass. The n-3 long chain polyunsaturated fatty acids content (AA, EPA and DHA) was almost double in these rabbits ($P<0.01$). Our study suggested that rearing rabbits in outdoor ark system could be a possible alternative housing system to improve the rabbit's welfare and some meat quality traits.

Key words: Rabbit, Arks, Meat quality, Oxidative status.

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Effect of environmental enrichment on carcass and meat quality in growing rabbits reared in collective pens

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To evaluate the effect of the presence and the type of environmental enrichment in different collective housing systems, 504 crossbred rabbits were weaned at 33 d of age and housed in 16 pens (1.68 m^2) with plastic slatted floor. Half of the pens were equipped with a plastic slatted platform (50 × 120 cm; 30 cm above the plastic floor). Within pens with or without the platform, half of the pens were additionally equipped with a plastic tube (ϕ 20 cm; length: 50 cm) for hiding. Within housing system, rabbits were slaughtered at 68 or 75 days of age. The presence of the platform increased slaughter weight (2495 *vs.* 2424 g; P<0.10) and the occurrence of rabbits injured at genitals (20.6 *vs*. 11.7%; P<0.01). Slaughter results were not affected, but dissectible fat was higher in rabbits kept in pens with the platform compared to those without (3.0 *vs.* 2.6%; P<0.01). Ultimate meat pH did not change whereas the lightness index of both *l. lumborum* (53.7 *vs.* 51.3) and *b. femoris* (50.7 *vs.* 49.8) was higher in rabbits reared in presence of the platform (P<0.01). The presence of the tube decreased slaughter weight (2403 *vs*. 2515 g; P<0.01) and reference carcass weight (1217 *vs.* 1276 g; P<0.10), but did not affect other carcass traits. The pH and colour of main muscles were not affected, but significantly higher shear force was measured on *l. lumborum* (4.63 *vs.* 4.30 kg/g; P<0.05) and *b. femoris* (3.15 *vs.* 2.83 kg/g; P<0.01) of the rabbits kept with the tube compared to those without. In conclusion, the presence of enrichment may affect both positively (platform) or negatively (tube) slaughter results and carcass traits in rabbits kept in collective pens. Further investigations are needed to ascertain the effects of the platform on aggression frequency.

Key words: Platform, Tube, Age, Slaughter results, Aggressions.

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Effect of feeding programme on performance and health of growing rabbits

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The effects of two feeding systems (L, *ad libitum vs.* R, feed restriction) combined with three diets (HH, a high digestible energy diet, 11.1 MJ DE/kg, during the whole trial *vs.* MM, a moderate DE diet, 10.7 MJ/kg, during the whole trial *vs.* MH, a moderate DE diet during the first three weeks and a high DE diet during the last two weeks) were evaluated in 252 commercial crossbred rabbits kept individually from weaning to slaughter (34 to 70 d of age). The restricted rabbits were fed from 85% of the theoretical *ad libitum* intake at the beginning of the trial to 100% of the *ad libitum* level at the end of the 3th week. During the first week, growth rate was impaired by feed restriction (53.6 and 46.8 g/d in L and R rabbits, P<0.001) because of the lower feed intake (107 *vs*. 93 g/d). During the second week, the appearance of digestive disorders in L rabbits reduced the difference in feed intake with R groups (134 *vs*. 129 g/d, P=0.05) and growth rate was similar in the two groups. During the third week, L rabbits had similar feed intake (151 *vs*. 148 g/d, P>0.10) and lower growth rate compared to R rabbits (48.2 *vs*. 51.3 g/d, P=0.05). During the fourth and fifth weeks, neither growth rate nor feed intake differed between the two groups. In the whole trial only feed conversion was improved by feed restriction (2.96 *vs.* 2.89 in L and R rabbits; P<0.01) and by the high-DE program (2.89 *vs*. 2.93 and 2.97, for HH, MH and MM groups, respectively; P<0.05). Morbidity was lower in the restricted group (5.5% *vs.* 2.4% in L and R rabbits; P<0.001) and in HH and MM groups compared to MH group (3.6% and 2.4% *vs.* 5.9% respectively; P<0.001). In conclusion, feed restriction improved feed efficiency and health, whereas the change from a moderate to a high energy diet during the growing period could impair rabbit health.

Key words: Feeding plans, Dietary energy, Growth performance, Morbidity.

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Using DNA microarray technique in determining the virulence gene profile of rabbit *Escherichia coli* strains isolated from colibacillosis outbreaks in Northern Italy

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A microarray targeting both the virulence genes related to known *Escherichia coli* pathotypes, the markers used in phylogrouping and genes encoding for somatic antigens used in serotyping of *E. coli* was applied in determining the virulence gene profiles of 26 rabbit enteropathogenic *E. coli* strains isolated from 16 colibacillosis outbreaks located in two regions of Northern Italy. *E. coli* strains were selected on the basis of the combined criteria related to farm origin, production unit, biotype, and the presence of the *eae* gene as detected by classical polymerase chain reaction. The distribution of virulence genes encoding for Locus of enterocyte effacement (LEE), LEE type III secretion system (T3SS), non-LEE T3SS translocated proteins and adherence factors was determined. All strains but one belonged to phylogroups A and B1 characteristic of nonhuman-associated diarrheagenic or commensal *E. coli* strains. One strain belonged to phylogroup D, which typically groups pathogenic *E. coli* associated with human outbreaks. A prevalent association between the O103 serotype *wzy* gene with the rhamnose-negative phenotype (biotype 12 or 14) was found. The most prevalent LEE profile found in the examined strains was *ler/tir-1/eae(beta)/espA-1/espB-3/escN/eprJ*. Positive signal for *cesT* and *espD*, detected by additional PCR test, was also found in all strains. Twenty-four strains possessed either the adhesive factor rabbit-2 (*afr/2*) or the plasmid Rabbit adherence locus encoding gene (*ral*), however, two strains were positive for an incomplete cluster of the plasmid Bundle-forming type IV pilus encoding gene (*bfp*). The *ral* gene was detected in all O145 strains while the *afr/2* gene was detected in all O103 serotypes. Finally, the combined or single presence of a set of LEE and/or non-LEE effector proteins encoding genes, namely *espG*, *cif* and *map*, was consistent with the severity of pathologic lesions found on the rabbits in the investigated outbreaks.

Key words: Colibacillosis, LEE, Microarray, REPEC.

On short term the reduction of antimicrobial consumption in fattening rabbits seems not correlated with decreasing antimicrobial resistance

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The antimicrobial resistance (AMR) is increasing worldwide and is currently a public health priority. The huge consumption of antimicrobials for prophylaxis or therapy of food producing animals (FPA) enhances the selective pressure on both pathogenic and commensal bacteria. The latter bacteria, especially when part of the gut microflora, work as reservoir of resistance genes that can be transferred to bacteria harmful for humans. Concrete actions are now solicited from international bodies (WHO, European Commission) to reduce antimicrobial consumption in FPA so to reduce the AMR overall burden. To evaluate the short time effect of an antimicrobial prudent use (APU) on the overall AMR rate in rabbit industrial farming we compared the minimal inhibitory concentration (MIC) of six antimicrobials commonly used to cure enteric disorders (enrofloxacin, colistin, apramycin, tetracycline, sulfisoxazole and trimethoprim-sulfametoxazole) against 297 *Escherichia coli* isolates collected in two rabbit herds over a three years' period. Rabbits of each fattening cycle were divided into two group: one treated with tetracycline with prophylactic doses added to the feed during all the fattening period (treated) and the other with no feed medication (not treated). Over the experimental period rabbits were kept in welfare conditions that included medication if needed and according to the antimicrobial susceptibility testing of isolates and to the APU principles. Results showed high level of AMR for all tested antimicrobials with no significant differences between strains isolated from treated and not-treated animals. We also did not record any trend in MICs values over three years. It seems that acting with APU principles in the fattening units only, might not be enough to detect reduction of AMRs and that the entire breeding system (i.e. the entire production pyramid) should to be involved in the AMR control policy.

Key words: Antimicrobial resistance, Rabbit, *Escherichia coli*, MIC.

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Antimicrobial consumption in industrial rabbit farming, Italy, 2010 to 2013

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After scientific evidence of critical levels of antimicrobial resistance in bacteria from humans and food producing animals, the European Commission calls for concrete actions at national level on the monitoring of antimicrobial consumption in food producing animals. In the 4th report of the ESVAC EMA project, Italy is among the EU members with the highest sale rates of veterinary antimicrobial drugs. Further, compared to other food-producing animals, the rabbit industrial farming was the production mostly exposed to consumption of antimicrobials during 2010-2013 in France (ANSES). To address the lack of information on the usage of antimicrobials in the Italian rabbit industrial farming, since 2010 we have been monitoring the consumption of antimicrobials in 32 rabbit farms of North-eastern Italy. The project is a pilot teamwork with the local Veterinary Public Health and farmers that volunteered to participate. Data are based on records of antimicrobials prescribed and purchased that were assumed to be consumed. Results showed 28% decrease of antimicrobial consumption over the 2010-2013 period (from 2681 mg/Kg LW in early 2010 to 1921.9 mg/kg LW in late 2013). Important differences between farms and drugs on consumption of antimicrobials were noticed. Overall we reported a decreased exposure to colistin (from 246 to 60 mg/Kg LW) and to tetracyclines (from 2035 to 1308 mg/Kg LW), but also an increased exposure to fluoroquinolones (from 3.7 to 20.9 mg/Kg LW), bacitracines (from 5 to 26.5 mg/Kg LW) and sulphonamides (from 146 to 273 mg/Kg LW). No changes in aminoglycosides, macrolides or pleuromutilins prescribed and purchased were observed in same period. Despite the overall decrease of antimicrobial consumption in the rabbit intensive farming we have been monitoring, more effective short term actions are required to align the antimicrobial consumption in rabbit meat production to the other food-producing animal species in the European Union.

Key words: Rabbit, Antimicrobial consumption, Antimicrobial resistance.

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Concurrent infection of rabbit (*Oryctolagus cuniculus*) and Italian hare (*Lepus corsicanus*) by Rabbit Haemorrhagic Disease type 2 virus

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Rabbit haemorrhagic disease virus (RHDV), member of the genus *Lagovirus*, causes rabbit haemorrhagic disease (RHD), a fatal hepatitis of rabbits. Similarly, a related Lagovirus, namely European Brown Bare Syndrome Virus (EBHSV) causes disease in hares. Until recently, the strict host restriction of the two viruses was unquestionable, but in the last years, a new RHDV-related virus emerged, called RHDV2, which was reported to cause RHD in rabbits and Cape hare (*Lepus capensis*). Here we report a case of RHDV2 infection in Italian hares (*Lepus corsicanus*), during a concurrent RHD outbreak in wild rabbits, in Sicily. The disease was fatal for 39 out of 40 rabbits, which lived in a fenced area about 200 m^2 wide. Simultaneously, an Italian hare died with typical signs of RHD. This animal was part of a group of 30 Italian hares, which were housed about 30 m from rabbitry. Necropsy findings and gross lesions were consistent with RHD, and RHDV infection was confirmed by negative staining electron microscopy, ELISA and specific RT-PCR. The latter targets VP60 gene of both EBHSV and RHDV, and it discriminates them according to the dimension of amplicons. The nucleotide sequence analysis of VP60 gene ascertained that the animals were infected by the emerging RHDV2, and that both rabbits and hare were infected by the same strain, as sequences were 100% identical among themselves. These findings were then confirmed by serological investigation, carried out by using a sandwich ELISA test with anti-RHDV monoclonal antibodies (MABs) produced against RHDV, RHDVa and RHDV2. A competition ELISA with MABs specific for EBHSV, RHDV and RHDV2 was then carried out to test sera collected from hares 3 months after the outbreak. Hares were seronegative for RHDV, RHDV2 or EBHSV. On aggregate, these findings highlight that RHDV2 may infect not only *O. cuniculus*, but also *L. corsicanus*. However, the latter species seems less susceptible to the virus, since the animals, which lived together with the affected hare, resulted seronegative and asymptomatic. On the other hand, it is evident that RHDV2 is potentially capable to infect different *Leporidae* species. This implies that circulation of RHDV2 among wild animals should be strictly monitored, as the infection of more widely distributed species (e.g. *Lepus europaeus*) may lead to a rapid global spread of RHDV2, also threatening vulnerable or endangered species.

Key words: RHDV2, *Oryctolagus cuniculus*, *Lepus corsicanus*, Host susceptibility.

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Dynamics of the serological response of commercial rabbits to Borghi and SG33 vaccine strains

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The effectiveness of live virus vaccines against rabbit myxomatosis is well established. Humoral response is not considered protective against the disease but represents a useful parameter to monitor the vaccination rate or the diffusion capability of wild strains in unvaccinated flocks.
The aim of this study was to evaluate whether differences in antibody titres may occur over the time in farmed rabbits vaccinated with the strains "Borghi" and "SG33". Three groups of 50 male rabbits aged 34 days, were housed and blood tested (1st sampling). Six days later, two groups were intradermically immunized with the commercial vaccines containing strain "Borghi" and "SG33". One group was kept as not-vaccinated control. Blood samples were collected at 12 (2nd sampling), 20 (3rd sampling), 29 (4th sampling) and 41(5th sampling) days post housing. Specific antibodies were detected by a commercial indirect enzyme-linked immunosorbent assay (ELISA) (Civtest CUNI Myxomatosis) and quantified in ELISA units (EU).
The ANOVA showed the mean of log-transformed ELISA values of vaccinated animals not differing significantly from values of the control group at 1st and 2nd sampling, but significantly higher starting from the 3rd sampling for group "SG33", and from the 4th sampling for the group "Borghi. The overall amount of antibodies in the group "Borghi" was higher compared to group "SG33" in 4th and 5th sampling. After applying the cut-off value of > 0.20 EU, a higher amount of immunised rabbits was detected in group "SG33" at 3rd and 4th sampling but not at 5th sampling when the number of positive rabbits was higher in group "Borghi".
In conclusion, strain "SG33" seems able to produce a faster serological response compared to "Borghi", but the latter yielded the highest coverage of immunised rabbits at 5 weeks post vaccination.

Key words: Rabbit, Myxomatosis, ELISA, Vaccine strains.

Antimicrobial susceptibility of clinical isolates of *Escherichia coli* in rabbit industrial herds in 2010-2014

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Clinical sampling, diagnostic on-site tests and susceptibility testing are top tools for strategies recommended by the European Council to tackle antimicrobial resistance (Council conclusions, 2012/C211/02). Enteropathogenic *Escherichia coli* (EPEC) provokes diarrhea in suckling and post-weaning rabbits that affects the rabbit industrial farming. Several risk factors can trigger EPEC overgrowth and cause outbreaks that require antimicrobial therapy so to reduce flock morbidity and mortality, and guarantee the welfare of animals. To support veterinary practitioners prescribing correct therapy we describe the outcome of five years (2010-2014) susceptibility testing (AST) of *Escherichia coli* from colibacillosis outbreaks occurred in rabbit industrial herds located in North-eastern Italy. Susceptibility was tested by disc diffusion against 13 antimicrobials (paromomycin [PAR], apramycin [APR], ampicillin [AM], flumequine [UB], gentamicin [GM], sulfonamide [S],sulfonamide-trimethoprim [SXT], tetracycline [TE], neomycin [N], 2nd generation quinolones [ENO], florfenicol [FFC], colistin [CL], amoxicillin and clavulanic [AmC] and 3rd gen. cefalosporine [CTX] in 724 isolates classified according to biotype. Interpretation was according to clinical breakpoints. Results showed high *in vitro* resistance to PAR (26% isolates resistant), APR (65%), AM (39%), UB (31%), GM (59%), S (94%), SXT (72%), TE (94%) and N (24%), moderate *in vitro* resistance to ENO (15%), FFC (11%), CL (8.7%), and low *in vitro* resistance to AmC (1.5%) and CTX (2.2%). Over the five years span a significant increase of *in vitro* resistance to ampicillin and amoxicillin (P=0.004), colistin (P=0.024), flumequine (P=0.016), florfenicol (P<0.001); the increase of resistance to 2nd generation quinolones (P=0.06) was close to the significance limit of 0.05. The majority (93%, 672/724) of tested isolates was resistant to at least 3 antimicrobials. 28% of isolates belonged to top four multi-resistance profiles (APR-GM-S-SXT-TE, APR-GM-S-TE, S-SXT-TE and PAR-AM-APR-GM-N-S-SXT-TE) and were detected in EPEC biotypes 12, 14, 28 and 30. Thus sulfonamides, tetracyclines, apramycin and gentamicin are mostly ineffective for rabbit EPEC.

Key words: Rabbit, *Escherichia coli*, Antimicrobial resistance.

Environmental impact of rabbit production through Life Cycle Assessment

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Greenhouse gas emissions (GHG) associated with agricultural activity are estimated to be 7.5% of total Italian GHG. Several studies have highlighted the environmental impacts caused by animal origin product, but studies on rabbit production are few.

The purpose of this study was to evaluate the impact of rabbit production system from cradle to farm gate on Global Warming Potential (GWP) performing through Life Cycle Assessment. Data were collected in a representative intensive rabbit farm. GWP (kg CO_2 eq) was quantified according to IPCC and Simapro software (8.0.3) was used to calculate this impact category. Land Use Change was included in the analysis in the case of soybean meal and soybean oil (imported from Brazil). The functional unit was 1 kg of live weight produced.

The value of GWP detected in this study was 5.54 kg CO_2 eq/kg live weight produced. This value is higher than those found in other monogastric species (2.2 and 3.3 kg CO_2 eq/kg for poultry and pig production). The largest contribution to GWP came from growing sector with 65.3%, while breeding sector (rabbit does and young female) showed less impact (21.6 and 13.1%).

The production of raw materials is the main process responsible for the GWP (78.1%). Soybean and alfalfa meal had the highest proportion of GWP of the feed components (23.6 and 7.3%). The high GWP of soybean meal is largely caused by transport of this raw material and by emissions released as a result of land-use changes. Housing and manure storage, instead, had less impact for GWP (12.7%).

In conclusion, feed production is the factor that contributes most to the GWP; it could be useful to pay attention to the protein level of the diets during growing phase and to moderate the use of soybean meal in order to decrease emissions in rabbit production.

Key words: Rabbit, Life Cycle Assessment, Global Warming Potential, Feed.

Susceptibility testing in *Escherichia coli* strains using the disk pre-diffusion, the disk diffusion tests and the minimum inhibitory concentration

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Colistin is a cationic polypeptide that binds to the outer membrane, leading to its disruption, mainly in Gram negative bacteria. In veterinary medicine, colistin is mainly used as oral preparation in order to treat infections caused by *Escherichia* (*E.*) *coli* strains, including rabbits. Even though the resistance to colistin in *E. coli* from a veterinary source have been seen occasionally, in the last few years various are the reports about the increasing resistance rate. In the diagnostic laboratory, the susceptibility test is performed using the disk diffusion (DD) method. Because of the scarce diffusion of the polymyxins in the agar, the accuracy of this test is under debate. The gold standard is represented by the minimum inhibitory concentration (MIC) test. Recently, authors described the pre-diffusion test (PDT) in order to differentiate susceptible from resistant strains in *E. coli* isolated from pigs. In our study 63 strains, isolated from rabbits affected by enteritis, were tested by the cited methods. The DD and the MIC were performed according to the Clinical and Laboratory Standards Institute (CLSI) guidelines. For the DD the interpretation was in accordance with Morales *et al* (2012). For the MIC, the interpretation was done in accordance with the EUCAST parameters. The PDT was performed as described by Boyen *et al* (2010) and the interpretative criteria of the obtained halo were as following: susceptible ≥15 mm; intermediate: 11-14 mm; resistant: ≤10 mm. In this study, different percentages of resistance have been obtained: 12.7% with the DD, 44.4% using the PDT, while only the 3.2% of the tested strains were resistant when submitted to the MIC. This study highlights that the DD and the PDT do not allow the discrimination between susceptible and resistant strains. In this scenario, only the utilization of the MIC could give accurate results both for therapeutic and epidemiological purposes.

Key words: *Escherichia coli*, Disk diffusion test, Minimum inhibitory concentration, Pre-diffusion test.

Survey on biosecurity measures and farmers' awareness about antimicrobial resistance in industrial rabbit farms of the Veneto region

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In comparison to other livestock, meat rabbits have fewer vaccines available for disease prevention. This aspect may explain the higher antimicrobial consumption and underlines the importance of biosecurity in this sector. Moreover, treatments are characterized by high frequency, long duration and oral route of administration, potentially resulting in underdosage in large groups of animals and contributing to antimicrobial resistance (AMR) development. In Italy, there is a lack of information regarding the application of biosecurity measures in rabbit farms and the level of farmers' awareness on AMR. This study aimed at filling this gap by means of a survey administered to 32 farmers randomly selected in the Veneto region. Part A of the interview collected information on holdings/management: synchronization of births, cage type, ventilation type, procedures of cleanliness, disinfection, etc. Part B investigated farmers' knowledge and concern regarding the correct drug usage and AMR issue. The results show that 75% of farmers were aged 40-60 years. Only half of the farms had either fences or barriers; 97% of farmers carried out at least one disinfection per each fattening cycle; all farmers applied rodent control. 75% of farmers did not apply "all-in-all-out" to fatteners, 85% has not practiced flock culling for the last five years and did not routinely subject newly introduced animals to a quarantine period. Although not having attended (86%) any education course on AMR, most farmers (90%) had knowledge on AMR, which in most cases (86%) was reported by their veterinarians/technicians. However, 80% of farmers did not know that some antimicrobials are critically important for human health and were not aware (60%) that antimicrobial usage in their farm might impact human health. In conclusion, these preliminary results indicate two criticisms in rabbit sector: a reduced generation turnover and a limited attention on the importance of biosecurity measures.

Key words: Survey; Rabbit farm; Biosecurity; Antimicrobial resistance.

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A prototype of colony cage with removable walls for improving the welfare of rabbit does: preliminary behavioural results

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Rabbit does are individually housed, but recently, on the basis of EU recommendations, great attention has been reserved for developing alternative housing systems (i.e. colony cage) closer to the living conditions of the wild rabbit. However, considering the great application difficulties of this kind of housing system, the aim of this study was to evaluated the effect of a new colony cage prototype (constituted by two separate floors for does and fattening rabbits equipped with removable walls) on the behaviour of rabbit does. To this aim, for two cycles, 30 nulliparous New Zealand White does were artificially inseminated and the pregnant ones were assigned to two groups: twelve does, were kept in single standard cages (group S) and twelve in three colony cages (group C). In group C the does were isolated by partition walls five days before kindling till one week after kindling; after weaning (30 days) does were moved in the colony cage on the upper floor, while young rabbits remained in group in the cage.

As expected, S does showed a more stable behaviour over time, while housing system interact on behaviour of C does in relation to different reproduction phases. Indeed, these showed a wider behavioural repertoire, as well as stereotyped and social behaviours that were not always friendly; in particular, these does showed higher incidence of aggressiveness during at the begin of the trial. Thereafter, an adaptation to establish hierarchies, dominance (15%) and lower *allo-grooming* (8,3%), was observed. Successively, the relations of mutual tolerance backed to normal levels.

The possibility to close the removable walls avoided problems related to litter exchange or aggression to other litters. In conclusion, the use of this cage prototype and consequently the possibility to modify social relationship along the reproductive cycle could reduce the incovenients registered in this kind of housing system.

Key words: Rabbit does, Colony cage, Behaviour.