Proceedings

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ECAWBM Annual Congress

and

IRSEA International Congress

Editors:

Patrick Pageat, Alessandro Cozzi, Cécile Bienboire-Frosini, Manuel Mengoli

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reface

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who will be presenting at it.

Two groups of people were particularly important in organising this meeting: the congress committee of the college and the local organising committee, headed by Patrick Pageat. The local organising committee faced not only the challenge of running a successful meeting, but also integrating it with the IRSEA (Institut de Recherche en Sémiochimie et Ethologie Appliquée) conference, which is now in its second edition.

Each international meeting has as its high point the paper presentation. I would like to acknowledge the work of the scientific committee for reviewing and selecting submissions. As you can say from the quality and range of the abstracts this is set to be another stimulating and successful meeting.

Any significant meeting cannot exist without the financial contribution of its sponsors. We should all thank for their kind and generous support.

Yours sincerely,

Jaume Fatjó
President of the ECAWBM

Preface

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President of the ECAWBM

Introduction

As President of the IRSEA (Institut de Recherche en Sémiochimie et Ethologie Appliquée) et Past-President of the ECAWBM (European College of Animal Welfare and Behavioural Medicine), it is a pleasure and a honour, to welcome you for this joined International Congress in the beautiful city of Apt (Provence, France).

I would like to thank the whole Scientific Committee, who has been in charge to evaluate and select the papers, which will be presented during this congress. This is a difficult and heavy work, which is crucial for the success of a congress, and I must say that the selected papers will, once again, confirm the reputation of the College Annual Congress, as well as the IRSEA International Congress.

I would like to underline the crucial support we have received from our Diamond Sponsor, Ceva Animal Health, which is providing its continuous and priceless help to our discipline. I would like to ask you to join us in our warm and sincere thanks to Ceva and to all their representatives in this congress.

This congress was not feasible without the unconditional and precious help of the City of Apt, led by its Mayor Olivier Curel. I thank him and all his wonderful Team for making such event possible, since 2010.

This introduction would not be really achieved, if I forget to present all my deepest feelings to the whole IRSEA Team, which has invested all its energy, time and skills, to organize the congress and create the special atmosphere which will make that you will just be expecting the 2016 edition of the IRSEA congress, when you will be on your trip to come back home.

Welcome to Apt, welcome to the ECAWBM-IRSEA congress, and thank you for visiting us.

Patrick Pageat

Thursday, November 20th 2014

ANIMAL WELFARE SCIENCE, ETHIC & LAWS.

ECAWBM and IRSEA joined session.



Oral presentations

Self-administration of ibuprofen in NMRI mice after vasectomy

E Clerget, M Kolb, J-P Dehoux.

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The aim of the present study was to assess the putative benefit of analgesia in mice following surgery (vasectomy), by measuring physiological and behavioural parameters. Importantly, analgesia was dispensed by free self-administration of ibuprofen dispersed in geldiet.

In the first experiment (#1), mice were allowed to choose between ibuprofensupplemented geldiet and non-supplemented geldiet. In the second experiment (#2), two groups of mice were tested for their consumption of geldiet: one group with an ibuprofen-supplemented geldiet and the other group with a non-supplemented geldiet. In both experiments, a video was recorded for 30 minutes on days: -1, 0 (surgery), +1, +2, +3, for offline analysis of behaviour. Mouse weight and temperature, as well as the geldiet(s) weight were also recorded.

Surprisingly, in experiment #1, the data showed that mice preferentially chose the ibuprofen-supplemented geldiet (consumption: 12.6 ± 1.0 g/j/mouse), even before surgery, while the consumption of non-supplemented geldiet remained low (consumption: 8.1 ± 1.2 g/day/mouse) (repeated measures ANOVA, F(1, 10) = 1.82, p= 0.031). In experiment #2, this result was confirmed since in mice receiving ibuprofen, the consumption of geldiet was higher (19.2 ± 1.0 g/day/mouse) than for the non-supplemented group (16.3 ± 1.0 g/day/mouse) (repeated measures ANOVA, F(1, 21) = 1.43, p= 0.05). In addition, experiment #2 showed that the self-selected "amount" of ibuprofen intake reversed some changes associated with post-operative pain as demonstrated by behavioural data.

The study demonstrated that self-administered ibuprofen provided through supplemented geldiet is an interesting method for alleviating pain in mice following vasectomy.

Dogs can be victims of Munchausen by proxy: a description of 12 cases.

Patrick Pageat - Invited Speaker

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Munchausen by proxy is a psychiatric condition, classified in the DSM-IV as a "factitious disorder." People affected by the disorder intentionally cause or attribute physical or psychological disorders to another person. The perpetrator's motivation is to assume the patient role by proxy in order to obtain the sympathy and attention of others. The disorder is commonly known by the name of Baron Munchausen, author of numerous travel and adventure stories. The most common victims of patients affected by "Munchausen by proxy" are their spouses, children, relatives living at home, or patients (when the affected person is a medical worker). The existence of animal victims of Munchausen by proxy was previously suspected. This paper describes twelve cases of dogs presented in different private and public veterinary clinics with various symptoms intentionally induced by one of their owners or keepers, between 1989 and 2006.

The dogs represented a typical population for the country of observation: there were 8 purebreds and 4 crossbreds, weighting from 2.5 to 31 kilos, 6 entire males, 1 neutered, 3 entire females and 2 spayed. They displayed a variety of symptoms; 9 dogs suffered from mild to severe digestive disorders (vomiting, gastric dilation, enteritis, colitis), 3 were affected by seizures, 5 had dermatitis, 2 had conjunctivitis, 4 suffered cardiac disorders and 5 dogs had a urinary disorder. Most of them displayed 2 to 4 different conditions, occurring separately but successively; as conditions improved through treatment, new unforeseen problems were diagnosed. When Munchausen by proxy was diagnosed, the disorders had been present for between 10 months and 4 years, and the dogs had been treated in 6 to 15 different veterinary clinics; characteristic of therapeutic nomadism. Retrospective analysis proved that moving to another clinic was induced by successful treatments. The professional profile of the perpetrators, as described in literature, included 5 nurses, 3 pharmacy technicians, 2 veterinary

technicians, 1 physiotherapist, and a sales representative for a pharmaceutical company. There were 8 female and 4 male perpetrators, which was also in accordance with the literature. All were very popular in the veterinary clinics where their dogs were treated, and were recognized as kind people and compliant owners who deserved warm support and sympathy.

Advancing animal welfare in veterinary education.

Titus Alexander, Nancy Clarke, Ellen Coombs, Joe Anzuino

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Through its Advanced Concepts in Animal Welfare (ACAW) programme, The World Society for the Protection of Animals works to support the inclusion of animal welfare within veterinary curricula worldwide, and many faculties have worked with us to integrate animal welfare into their teaching. We currently work with over 450 tertiary educators from universities in over 20 countries across the Latin America, Africa and Asia Pacific region.

Our main teaching support tool *Concepts in Animal Welfare* (CAW) was created in 2004 in collaboration with the UK's University of Bristol, and updated in 2007. The new 3rd Edition, launched in 2013, is designed to reflect the changing face of veterinary practice in relation to animal welfare.

With the OIE's recommendations that animal welfare is a foundational aspect for 'Day 1 Competencies', CAW is designed to enable teaching staff and students to access animal welfare science and apply it to their work. The resource contains up to date information on the latest animal welfare science; some existing modules have been expanded to include further information, and there are new modules on the role of vets in Disaster Management, and introductions to the OIE.

The ACAW programme and CAW resource approach to teaching and learning can enhance the student experience and ensures that the objectives for learning about and understanding the importance of animal welfare are integrated into veterinary training, including by lecturers who are new to animal welfare. This poster details the current global approach of the ACAW programme.

Improving equine welfare: guidelines on the management on the management of horses kept in research facilities.

V Jonckheer-Sheehya, Katherine A. Houptb

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Horses and ponies used for research are typically housed in individual stalls with little or no opportunity for social contact with conspecifics. This type of management system severely compromises the welfare of horses as isolated subjects show behavioural and physiological stress reactions that may also negatively influence experimental data collected from them.

Group-housing horses is well recognised as the best method to fulfill the physical and behavioural, especially the social needs of horses, as well as having a positive influence on horse–human interactions during training. Furthermore, some feel that it is not morally acceptable to house horses individually.

There are various methods of group housing horses such as in communal barns, individually tethered with no partitions between them in large sheds, at pasture or in multi-functional area group housing systems which have different functional areas including an exercise area that simulate the activity of horses within a group by careful spatial organisation of the roughage and concentrate feeding areas, the watering station, the rest area, the rolling spot and the exercise area. Examples of these multifunctional group housing systems include the Paddock Paradise system and the computerised, automated Hit Active Stable®. Aggression is often a concern; however, in correctly set up systems this is not a major problem. The advantages and disadvantages of different types of group housing systems will be discussed with guidelines on how to set up the optimal equine housing system in the research setting.

Animal-based measures used as a diagnostic tool for the welfare of dairy cows on farm.

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Using the Welfare Quality® protocols as a starting point, a practical approach was adopted to identify the critical points on a farm and establish some prevention strategies. The aim of this report is to describe such diagnostic approach on dairy farms. Three cases, visited in Spain, are presented in this report. The first farm was considered as "conflictive" by its assurance company due to the high percentage of downer cows. The day of the visit, lameness and cows' dirtiness were detected as two main animalbased issues. The mean flight distance obtained at the human-animal relationship test was not considered as problematic; however, a high percentage of cows could not be touched by the assessor. The second case presents a farm where the farmer asked for advices on the welfare state of its cows related to changes he was doing in the facilities. The day of the visit, a high percentage of animals presented severe lameness as well as swellings and lesions on the medial part of the hock. Moreover, many cows were resting partially outside the lying area. The third farm is part of a farmer association interested in assessing and improving the welfare of its animals. The day of the visit, many animals were panting. Social negative interactions were especially frequent at the drinker areas. Heat stress and prolonged thirst were two major related welfare concerns. After sharing the results with the vet and farmer responsible for each farm, a presumptive diagnostic was made and some corrective measures were proposed on the management, resting and milking facilities, and water supply on the three farms, respectively.

Is cloning of animals ethical?

Madeleine L.H. CAMPBELL

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In 2012, the Federation Equestrian Internationale rescinded a rule banning cloned horses or their offspring from competing. In 2013, the European Commission announced proposals for a ban on cloning of farming animals (except for research, conservation, and pharmaceutical/medical reasons) 'to address animal welfare and other ethical concerns'. Thus a situation may shortly exist in Europe whereby cloning of farming animals is banned for some but not all purposes, whilst cloning of non-food producing horses and smaller companion animals is fully permitted.

This paper investigates whether such a discrepancy in regulation is supportable ethically. It will analyse whether public sentiment that animal cloning is 'a step too far' can be justified on welfare or moral grounds, for all or any animal species, and for all or any purposes.

Many of the moral reasons for banning cloning in humans do not apply to animals. However, adverse outcomes across generations mean that cloning of animals (currently) carries significant welfare costs, which are ethically important. Where animals are being used for the same purpose, legal discrepancy allowing cloning of some species but not others could only be justified by evidence that welfare problems which are known to occur in some species do not occur in others. Such evidence is currently lacking.

In a cost:benefit analysis, the only ethical reasons for cloning using current techniques are pharmaceutical/medical ones. Although cloning is feasible, until welfare costs can be reduced the cloning of all animal species for other reasons is further than we should go.

The ethics of novel veterinary therapies: Innovation, Research and

Communication.

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Novel veterinary therapies may help individual patients and advance the body of

veterinary medical knowledge, but may also cause significant suffering without

sufficient benefits. Relevant clinical decisions are not subject to any effective regulatory

control to safeguard welfare, in comparison to legal frameworks and protocols for

scientific procedures (e.g. EU Directive 2010/63/EU].

The purpose of this work was to identify how adapting research ethics frameworks can

help develop clinical ethics frameworks regarding novel therapies.

Relevant literature was reviewed within human medical ethics and animal research

ethics. Key items were modified for veterinary ethics, including through substitutional

transpositions, contextualization and adaptations reflecting reported human-animal

differences.

Key proposals for clinical ethics of novel therapies identified include:

Avoiding potential conflicts of interest

Prior full comparative literature searches to assess expected utility, alternatives and

potential refinements

Considering species-specific biology and welfare needs

Describing the treatment, including expected outcomes as hypotheses, potential hazards

and treatment categorisation

Full, explicit and informed client consent or context-appropriate alternatives

Predetermining

pain and suffering assessment protocols

severity limits

outcome-based treatment success criteria

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decision-making limits, e.g. performing single major surgery
contingency plans including humane endpoints
Individually tailored analgesia
Training clinicians and owners
Systematic progression from pilot treatments to wider applications
Baseline treatment of all patients (i.e. no undertreated control patients)
Extensive record-keeping and audit, in particular for unintended consequences
Publishing results according to adapted "ARRIVE" guidelines.

Preliminary study of hospitalizes horse welfare and creation of score of quality of life of the hospitalized horse.

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Many studies deal with the welfare of hospitalized companion animals, but very few pertain to horses. The aim of this study was to assess well-being in hospitalized horses. A series of parameters were monitored in 10 horses aged 8 months to 14 years admitted to the equine hospital for colic: 5 were treated through surgical intervention, while the other 5 received medical treatment. Physiological indices included continuous heart rate, cortisol plasma concentration and pain score. Behavioral parameters were used to help establish a quality of life score. This score also included other factors such as free access to food, interaction with others horses, and the number of negative events per day. Additionally, a behavioral "profile" (aggressive, cooperative, anxious, indifferent, annoyed) was established for each horse based on a compilation of behavioral parameters, and the aversive grade of external events from the horse's point of view was assessed.

For 90% of horses, quality of life improved throughout hospitalization, and this was associated with a reduction in cortisol plasma concentration in 88.9% of horses. Plasma cortisol was slightly higher in horses treated surgically (with a mean of 156.1 nmol/L) compared with horses treated medically (mean of 149.9 nmol/L), possibly due to an overall diminished well-being.

62.5% of horses became "cooperative" on the first day of re-feeding during recovery. Among the events performed during medical follow-up, transcutaneous ultrasound was surprisingly found to be the most aversive act across all of the horses studied.

This study offers a practical approach to the investigation of welfare during the hospitalization of horses, the preliminary results of which warrant further research.

Assessment of Risk to Animal Welfare During Slaughter Without Stunning; Monitored Using Clinical Reflexes as per EFSA "Toolbox" (Council Regulation EC 1099/2009) and the Amelioration of this Process.

John CRANLEY

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Clinical (CNS) reflexes as per the EFSA "Toolbox" were used to assess the onset of insensibility in non-stunned slaughter in poultry, sheep and cattle. Consciousness was assessed using corneal, pupillary, nictitating reflexes, mouth movements and muscle tone. The time from incision to insensibility was measured using a digital stop-watch, the reflexes were tested using a LED or pencil torch light, and with touch testing corneal reflexes. Clinical studies were carried out recently, with ethical approval, under abattoir conditions. The slaughterers were experts, using sharp blades.

Non-stunned calves took longest to lose reflexes post incision. In a study of 100 calves the average time to loss of reflexes was 2minutes post incision. One calf took 6 minutes and another 5 minutes to lose all reflexes.

In 800 non-stunned lambs, reflexes were found to take up to 65 seconds to disappear. Evidence of resurgence of reflexes was found, in lambs and calves hoisted after cradle restraint.

In 1774 non-stunned broilers, an angry bird reflex was followed by loss of reflexes between 45 and 65 seconds post incision, with 0.3% showing reflexes at 150 seconds post incision.

The absence of reflexes is the legal requirement for the assessment of insensibility before release of the animals from restraint. Non-stunned creatures were conscious post incision. Therefore evocation of reflexes risks triggering an angry bird response. Restraint until death avoided this response. Equally, delay in hoisting sheep and lambs until death, avoided any risk of resurgence of sensibility.

Improvement of coping strategies in horses (*Equus caballus*) thanks to long-term memorisation

<u>Tiago Mendonça</u>^{a,b}, Alessandro Cozzia, Patrick Pageata, Céline Lafont-Lecuellea, Philippe Monnereta, Gonçalo da Graça Pereirab, Manuel Mengolia

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Different studies in animals show that cognition and memorisation are essential to enable the animal to adapt to changes in its surroundings. The horse creates strategies to cope with new situations, promoting its welfare.

This study was designed in order to analyse long-term memorisation and recall capabilities in horses through a cognitive test.

Using a two-figure device, 16 horses were involved in a cognitive test: 11 horses had passed the test a year earlier (experienced horses; EH) while 5 new horses (naive horses; NH) were attempting it for the first time. Horses had the possibility to choose between two geometrical figures (a triangle and a circle) in order to obtain a reward: the circle always corresponded to the correct choice. The number of correct, incorrect and 'out of time' choices (horses had one minute to choose) was recorded, with each test lasting 10 minutes per horse.

The results showed a significant difference in the number of correct choices between EH and NH (Mann-Whitney U test, p<0.01; EH: median=14; NH: median=2). There was no significant difference regarding the number of mistakes (Mann-Whitney U test, p=0.50), however, there was an important variation in the 'out of time' responses (Mann-Whitney U test, p<0.01; EH: median=0; NH: median=6).

These results demonstrate long-term memorisation and recall abilities in horses, resulting in increased performance when reliving a positive experience through efficient coping strategies. It is crucial to link these abilities with the adaptation process in order to improve our understanding of horse welfare.

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A burdensome task: Setting thresholds for evidence weight in animal welfare science.

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Animal welfare science provides an intellectual authority that has helped drive policies and changes in practice. Policymakers or industry bodies may require a weight of evidence that a practice is harmful before any change is implemented, such as banning unenriched battery cages for poultry, electronic shock collars for dogs, wild animals in circuses or trade in wild species as pets. This burden can avoid inconstancy and some errors, but can risk unintended consequences including lack of progress, delays, skewed research funding priorities or tacit promotion of conservative or liberalist philosophies. The benefits and risks depend on how high the bar is set.

The purpose of this work was to develop an approach for setting burdens of proof *a priori*, in order to inform research priorities and project design, drawing on subjective utility theory and Bayesian theory.

A priori burdens of proof should be based on (a) a prior evaluation of the degree of harmfulness (based on intensity, duration and frequency) and (b) the prior confidence that a state constitutes poor welfare (e.g. based on an expert panel view). These can be used within a precautional approach as prior inputs for Bayesian inference. Animal welfare scientific studies may then be (a) not required, (b) deemed never able to satisfy an insurmountable burden or (c) required with a particular burden of proof. This last may be explicitly expressed as a particular effect size or likelihood ratio, allowing animal welfare science to follow other scientific fields such as ecology in less unthinking reliance on p-values.

Using semiochemicals to protect livestock species against insect bites: a new promising and sustainable strategy

<u>B. Creton</u>, A. Cozzi, M. Robejean, J. Leclerc, D. Saffray, H. Barthelemy, E. Codecasa, C. Lafont-Lecuelle, P. Bursztyka, L. Bougrat, P. Pageat.

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Due to overcrowding, farm animals are often subject to epizootics causing significant economic loss and welfare concerns. Insects are significant vectors of pathogens for different species, including humans and livestock animals. It's therefore relevant to develop a non-toxic strategy to control insects, thereby preventing disease. Today, due to global warming, health authorities of western countries fear an increase of vector diseases (African Horse Sickness, bluetongue). The aim of this study is to assess the protection efficacy offered by a semiochemical applied on sheep against an aggressive form of diptera, *Aedes aegypti*.

A randomized and blinded paired-samples study design was carried out using 12 sheep. Two 15 cm areas were shaved on each sheep's shoulders. These areas randomly received a placebo or semiochemical treatment (randomized and stratified especially concerning laterality). The areas were then exposed for 30 minutes to 50 caged female *Aedes aegypti*. Assayed insects were killed and engorged insects were counted.

The median percentage of gorged females was 70% for the control treatment and 1% for the semiochemical treatment, indicating a significant difference between treatments (Z=3.06; p=0.002, Wilcoxon for paired-samples test). The tested semiochemical provided 94% protection against mosquito bites for a period of 30 minutes.

Further assays need to be performed on other farm animals and other pathogen vectors, for example Culicoides, to verify efficacy against the transmission of different diseases. This preliminary data highlights the potential interest of a semiochemical approach in preventing hematophagous insects from attacking farm animals.

Social organization of three groups of Beagles permanently locked in a semi-open laboratory kennel.

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The hierarchical social organization has long been applied to dogs by extrapolation of observations made on their ancestors, the wolves. The current debate challenges hierarchy and leans more towards a dyadic organization. Social organization of laboratory dogs housed in human imposed social group has never been studied.

Interactions between a group of four females and two groups of three males were observed during 60h: type of interactions (negative, positive, neutral) and distance between dogs were recorder every 5 mn by individual during 1h with the focal animal sampling method.

No specific pattern could be demonstrated. Social interactions differ among individuals (T Test, p-value < 2.2e-16) and among groups. Some dyads interact a lot and others don't interact at all. Some dyads display mostly positive interaction and others essentially negative. Dogs interacting the most are the closest (Chi-2 test, p-value < 0.05). David's score failed to detect any dominance relationships (David's score between 0.4 and 0.6) for all dyad except one. Size of the kennel has an impact on dog interactions. Dogs in smaller kennel display fewer social interactions most stereotypies and most aggressive behaviors and vice versa.

We found that social organization of laboratory dogs maintained in human imposed social groups is similar to feral dogs groups. No linear hierarchy is observed. The group is non cohesive and results from an aggregation of dyads with specific interactions thought to be based on individual affinities. The environment seems to modulate the quantity and the quality of social interactions.

Horse whisperers and horse breaking: does the training method influence later horse welfare?

Chiara MARITI

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Horse breaking is traditionally based on restraining and preventing avoidance or escape.

So-called horse whisperers propose a different approach.

The aim of this research was to assess if the kind of training influences horse welfare in

later life.

Twelve unhandled, free-ranging female horses (2.5±0.5 year old) were equally divided

into a group trained with a gentle, traditional method in stables (TR) and a group trained

with a method inspired by horse whisperers in a round pen (ET). Horses' behavior and

heart rate after the training was assessed in an arena test (response to a novel

environment), a person test (stranger) and a grooming test (handling and grooming).

Groups were compared using a Mann-Whitney and t-test (p<0.05).

In the arena test, the TR group was more explorative (U=0.000; p=0.004) and had a

lower average heart rate (t=2.086: p=0.070).

In the grooming test, the ET group had a lower average heart rate (F=6.23: p=0.037)

and spent statistically more time with lateral ears (U=4.500; p=0.026) and a still tail

(U=5.000; p=0.041), while TR displayed more avoidance (U=3.000; p=0.015) and was

more difficult to groom. Moreover ET showed more curiosity to the stranger in the

person test (U=4.600; p=0.029), while heart rate was statistically lower (F=9.22:

p=0.013).

The horse whisperers' method had a negative influence on the response to a novel

environment, maybe due to the lack of familiarity with an indoor environment.

However, it also had a positive effect on the horses' response to man. The latter result

may have important consequences on the way horses perceive and experience mankind,

and therefore on their welfare.

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Welfare in dairy cows: the efficacy of Bovine Appeasing Pheromone (BAP) on milk indicators during lactation

Maria-Cristina OSELLA

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The aim of the study was to evaluate the effects of Bovine Appeasing Pheromone (BAP) in *Valdostana Pezzata Rossa* dairy cows in lactation during changes in housing and management conditions (move from indoors to outdoor pasture), usually presented as a highly stressful event.

The experimental design was a blinded trial against placebo in 2 parallel randomized groups with repeated measures. Thirty cows were enrolled and randomly divided into 2 groups: BAP (n=15) and placebo (n=15). The two groups were separately housed in the same farm and managed outside in two different pens at a distance of 200 m. The treatment and the placebo (5 ml) were poured on the inter-cornual area of each cow at the feeding rack weekly for 35 days (from T0 until T4). Parameters regarding the quantity and quality of milk production were measured at T0, T1, T2, T3 and T4: daily milk production (kg/day); somatic cell count (thousands of cells per milliliter); urea (mg/dl); proteins, fat, fat-free dry matter, casein, and lactose (%).

The results showed the effect of treatment over time on different milk indicators (mixed model ANOVA): daily milk production (df=4; F=2.64; p=0.037), somatic cell count (df=4; F=2.51; p=0.046); urea (df=4; F= 23.94; p<0.001); proteins (df=4; F=4.31; p=0.003), fat (df=4; F=5.6; p<0.001); fat-free dry matter (df=4; F=3.47; p=0.010); casein (df=4; F=4.6; p=0.002); lactose could not be analysed.

The use of BAP in dairy cows appears to be useful in improving the quality and quantity of milk and for managing a stressful situation in order to facilitate the natural process of adaptation during lactation under new outdoor conditions.

Development of the 'Ten Steps' Model to Solving Ethical Dilemmas in Practice.

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Veterinary surgeons regularly experience difficult ethical dilemmas, which present conflicting options and psychological pressures, and provide a source of stress. Resolving these dilemmas requires appropriate ethical consideration, communication and implementation of solutions. Such ethical decision making is an essential 'day-one skill' but veterinary practitioners lack formal guidance on decision-making, and instead rely on gut-instinct or unthinking compliance with requests or conventions. Ethics teaching similarly lacks recognised frameworks, leading to inconsistency or curricular omission.

The purpose of this work was to develop a practically-applicable framework for ethical decision-making and communication.

Relevant literature was identified within pure and applied ethics fields, categorised into normative theories (e.g. classical act-utilitarianism) versus procedural approaches (e.g. the Ethical Matrix). End-user needs were identified through literature searches regarding prevalent ethical dilemmas and the ethical skills and understanding perceived as being required. An overarching framework was modelled based on the Calgary model of communication and steps proposed by the extant literature.

The final model involves ten steps of:

- 0: Preparing
- 1: Describing the options available
- 2: Identifying Factual Influences
- 3: Identifying Ethical influences
- 4: Determining Principles

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- 5: Weighing the ethical-influences
- 6: Discounting prohibited options
- 7: Identifying the best option
- 8: Satisfying prerequisites
- 9: Acting
- 10: Reflecting and preparing for next time.

The framework was refined though informal discussions with practitioners and use in undergraduate and postgraduate teaching and will be presented in more detail.

The Pig Appeasing Pheromone: how chemical communication improve the welfare of pigs.

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Stress-related problems are very common in pigs and lead to impaired performance and increased vulnerability to infection at all stages of the animal's life cycle. Since the appearing pheromone has been identified in pigs, different studies have examined the efficacy of this semiochemical. Indeed, the use of pheromones is an interesting strategy to improve animal welfare in pigs.

The goal of this review was to create an inventory of all of the parameters that have been used to measure welfare in pigs and to highlight those that have been most successful in proving the effectiveness of Pig Appearing Pheromone in improving welfare. Data are focused on three categories of indicators: behavioural, zootechnical and physiological.

Several studies have proved PAPs effectiveness in decreasing stress-related behaviours; PAP is especially effective in reducing aggressions demonstrated by a reduction in the number and duration of fights during the weaning period, the fattening period and the group-housed pregnant sow period. Some studies have shown that PAP may be used to optimize growth after weaning with improvements in the Daily Weight Gain and the feed gain ratio. Moreover, concerning zootechnical parameters, one study reported greater weight homogeneity in pigs at the time of slaughter. Despite promising evidence, few studies have explored the physiological parameters. One such study demonstrated a dramatic decrease in the release of salivary cortisol during a social challenge between sows.

PAP is a modulator of homeostasis in individuals. The appeasing pheromone family, as shown in other mammal species allows the individual to cope with the environment and facilitates the adaption process.

Detection of enclosure shortcomings using a protocol to assess animal welfare in captive Dorcas gazelles (*Gazella dorcas*)

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Providing good standards of welfare to wild animals kept in captivity is important for both ethical and legal reasons. However, there is a lack of protocols designed for the assessment of welfare, which should be a major concern for centres and institutions that keep wild animals.

The aim of this study was to develop a protocol for the assessment of welfare in captive Dorcas gazelles (*Gazella dorcas*). The protocol would account for the following: the available biological literature on this species in the wild, the husbandry guidelines, particularly those developed by zoos for this species, and the protocol for the assessment of welfare in cattle from the Welfare Quality® project.

The protocol developed for Dorcas gazelles included animal and resource-based indicators such as body condition, water provision, enclosure size, climatic condition, lameness and integument alterations, expression of social interactions like affiliate behaviours and intra-specific aggression, group size and composition, stereotypies, environmental enrichment and medical training of the animals.

This protocol was then applied to five separate groups of gazelles from three different zoos.

The protocol was used to detect some areas for improvement of the enclosures where Dorcas gazelles were kept. Validation of the protocol is pending.

Poster presentations

Ethical considerations regarding the use of Guinea pigs to maintain uninfected mosquitoes.

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Hematophagous arthropods are an important source of discomfort for animals and humans. Furthermore, these bloodfeeding animals can transmit microorganisms such as bacteria and viruses. These microorganisms can lead to serious health problems (malaria, yellow fever, dengue, onchocerciasis, West Nile Fever, Dirofilariose, Leishmaniasis, bluetongue ...) and significant economic loss. Among bloodfeeding insects, mosquitoes are the main cause of vector borne diseases. The aim of this study is to describe an ethical approach to maintaining mosquito colonies for research purposes using guinea pigs (Cavia porcellus) as a blood source. The scientific study of mosquitoes improves our understanding of these vectors, enabling man to develop strategies to fight against the spreading of pathogens or to develop new repellents, insecticide molecules, or semiochemical treatments. The study of these bloodsucking insects requires large quantities of individuals readily available at all times. Laboratory breeding is therefore a necessity. Mosquito breeding methods differ according to the species bred and the breeding laboratory. Blood meals are constantly required to maintain large cohorts. The use of an apparatus to achieve artificial blood meals is less efficient than the use of live animals and impossible with certain species of mosquitoes. This is why many scientific teams favor the use of animals, however, in accordance with experimental ethics and legislation, using animals to maintain mosquito colonies must follow the ethical rules and principles for the inclusion of animals in scientific research.

Physiological response to repeated predation by piscivorous birds in Rainbow trout (*Oncorhynchus mykiss*)

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Farmed salmonids are subjected to numerous externals stress factors, such as predation by piscivorous birds. Following a single predator attack, fish display anti-predator behaviours and physiological stress, such as an increase in heart beat rate. Despite this clear evidence, there is a lack of knowledge regarding the influence of repeated predation on physiological stress levels in the rainbow trout.

In the present study, *O. mykiss* experiencing high repeated predation by piscivorous birds (Herons, cormorants and seagulls) on an inland aquafarm were divided into two groups. The experimental group (n=2) was subjected to a predator attack simulated by a plastic heron and was compared to a control group (n=2) without predation risk. The plasmatic cortisol level and the hepatosomatic index were calculated before, 1h and 3h after the simulated attack.

There were no significant differences between the attacked and control groups in either plasmatic cortisol levels or hepatosomatic index scores prior to and following the simulated attack. Data on plasmatic cortisol rate (range from 31.47 ± 22.39 to 66.32 ± 17.43 ng.ml⁻¹) and the hepatosomatic index (range from 1.05 ± 0.12 to 1.50 ± 0.20) can be considered as low, reflecting an unstressed state in the trout.

Previous publications and our preliminary results suggest the presence of a habituation process regarding repeated predator attacks in salmonids. This adaptive phenomenon allows fish to cope with the predation pressure without consequences to their vital functions, such as growth and reproduction. Beyond the direct consequences of mortality and injury, predation in aquaculture does not seem to have any additional repercussions on salmonid development.

Heterospecific Communication - Is Mutual Understanding Possible?

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Research devoted to heterospecific interaction between humans and pets is described.

The study of interaction of 132 cat and dog owners with their pets shows that despite

the apparent variety of words correctly perceived by dogs and cats, all these words can

be classified on two grounds: agents of action (the animal and family members), and

motivations to action with agents and objects, where the object and action form a single

syncretic unity. The animal's ability to differentiate the signals given by their owner

depends on the situational context. Correspondingly, we could say that animals create

mental representations of certain resistant integral complexes of the objects and

phenomena in their environment, but separate objects and phenomena that are included

in these complexes generally do not trigger a behavioral complex, inciting the owner to

take action. In the process of heterospecific communication, humans must reduce their

ability to differentiate meanings and "return" to the global undifferentiated state of the

early stages of mental development. The following methods were used in the research:

focused interviews and observations of pet and owner behavior during the

communication process.

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Shelter Quality: Welfare Assessment Protocol for Shelter Dogs applied in three shelters

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The Shelter Quality protocol was developed to provide a valid, reliable and practical tool for assessing dog welfare in long-term shelters. This protocol was built around the four welfare principles (good feeding, good housing, good health, and appropriate behaviour) described by the Welfare Quality® project for on-farm assessments of livestock species (Welfare Quality®, 2009). The aim of the present study was to test the protocol and to critically review the collected data.

Audits were performed in three different long-term shelters located in the same geographical area. Applying the method, scores were obtained for shelter level (general information, social housing, exercise, surgeries and control of pain, mortality, morbidity, feeding, emotional state); pen level (space allowance, bedding, sharp edges, water supply, diarrhoea, thermal comfort, barking level, abnormal behaviour, evidence of pain), and individual level (reaction toward humans, body condition, cleanliness of the animals, skin condition, lameness, coughing). Scores were recorded and analysed. Emphasis was placed on animal-based measures (also called outcome or performance measures) in an attempt to estimate the actual welfare state of the animals in terms of, for instance, their behaviour, health, and physical conditions. Results have indicated some applicative concerns: (1) the time required for auditing, (2) scoring difficulties, (3) the impossibility of using this method to identify problems and provide advice or treatment.

Welfare assessment in long-term sheltered dogs requires a different approach that involves the use of more than just standard indicators, especially when the shelter meets all criteria of existing legislation in the field.

Friday, November 21st 2014

BEHAVIOURAL MEDICINE. ECAWBM and IRSEA joined session.

Oral presentations

Anxiety-Reducing Effectiveness of a Therapeutic Calming Diet using a Model of Fear and Anxiety in Cats.

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The purpose of this study was to evaluate the effect of a diet supplemented with l-tryptophan and alpha casozepine, a milk hydrolysate, on cats identified as fearful.

Cats that were fearful of humans on home room assessment were significantly more inactive in both open field and human interaction tests compared to non-fearful cats. Furthermore we demonstrated that diazepam counteracts behavioral suppression and increases activity.1 Based on baseline testing 24 fearful cats were divided into two equivalent groups fed either test (Royal Canin Veterinary CALM Feline) or control diet (Nestle Purina Cat Chow Indoor Formula) for 4 weeks. At weeks 2 and 4 subjects were reassessed on both open field and human interaction tests. Data was analyzed using repeated measures ANOVA and the Fisher exact test for direct post hoc group comparisons.

On open field, for distance travelled, control animals showed a statistically significant decrease in activity at 2 weeks (p=0.023) and marginally significant decrease at 4 weeks (p=0.052). By contrast, the subjects on test diet increased their movement, but differences were not statistically significant. For inactivity duration, the control group showed a statistically significant increase at 2 weeks (p=0.042) and marginally significant difference at 4 weeks (p=0.052). By contrast the group fed test diet showed a statistically significant decrease at 2 weeks (p=0.0397) and marginally significant decrease at 4 weeks (p=0.0675). There were no group differences in the human interaction test.

These results demonstrate that a diet supplemented with L-tryptophan and alphacasozepine was effective in reducing anxiety in fearful cats.

Supplementation of polyunsaturated fatty acids, magnesium and zinc in dogs with behavior problems: Results of a pilot study

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Recent discoveries have shown that the chances of a dog developing a behavior problem may depend upon a number of factors including nutrition. The current pilot study was designed to provide a preliminary assessment of the efficacy of a dietary supplement containing omega-3 and omega-6 fatty acids as well as magnesium and zinc on some common behavior problems in a population of Iranian domestic dogs. In total, 42 dogs (17 male and 25 female) referred for behavioral problems, were given 330 mg omega-3 eicosapentaenoic acid, 276 mg omega-6 gamma-linolenic acid, 133 mg of magnesium, 5 mg of zinc for 6 weeks. Data were obtained using a questionnaire that dog owners were invited to fill out before and after the supplement treatment period. The questionnaire asked owners whether their dog had exhibited any of the six common behavior problems namely, excessive activity, inappropriate elimination, fearfulness, destructiveness and aggression towards unfamiliar people and dogs, on a 4- point Likert-like scale ranging from 0 (never or rarely) to 3 (very often).

After 6 weeks of consumption of the supplement most dogs showed a significant reduction in the median score for the severity of three behavior problems namely, fearfulness, destructiveness and inappropriate elimination. There were no significant differences in the median score for the severity of excessive activity and aggression towards dogs and unfamiliar people during the course of the study.

At present, few studies have been conducted to evaluate the role of nutrition in canine (problem) behavior. Studies that explore this relationship may help to improve the welfare of dogs and their owners.

Are nasal stem cells a promising approach in geriatric veterinary medicine?

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Age-related disorders (disorientation, disturbed sleep-wake cycles and loss of housetraining) commonly affect dogs and may lead to premature euthanasia. Standard methods of care for aging dogs are usually unsuccessful, as patients are often affected by several illnesses and have difficulty maintaining lifelong treatments, which leads to their discontinuation. In this context, stem cells are emerging as interesting tools in new therapies due to their multipotency and high proliferation. Nasal stem cells identified in the olfactory mucosa represent promising candidates for autologous grafts. Earlier, we showed that human nasal stem cells transplanted in a rodent model of amnesia could survive in injured brain areas and promote learning and memory recovery.

We have recently developed a new technique to collect, purify and culture canine olfactory stem cells obtained from one aging dog (16 years old) with success. This result allowed us to repeat the procedure with another dog (17 years old) displaying severe cognitive disorders. Before the biopsy, complete medical, neurological and behavioral analyses were performed. Twenty-five million olfactory stem cells were then amplified in culture over eight weeks and injected into the cerebrospinal fluid in the cerebromedullary cistern. There were no unwanted side effects caused by the biopsy and the cell graft in this dog. After a period of six weeks, there was a significant reduction in emotional and cognitive disorders, particularly on compulsive motor control (circling). We also observed improvements in exploration and the capacity to focus attention. These preliminary results suggest that stem cell therapy could be a promising approach in veterinary medicine.

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Epilepsy and behaviour in dogs: what is the link?

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Epilepsy is the most common chronic neurological disorder in dogs. Genetic epilepsy is widespread among dog breeds with an incidence of 5 to 20% depending on the breed. In human medicine, an increasing number of studies have identified neurobehavioral or psychiatric comorbidities associated with recurrent seizures disorders. There is a lack of data concerning dogs, but recent studies suggest that the same comorbidities exist in this species.

In a study of 80 dogs with idiopathic epilepsy, an increase in the behavioral factors such as fear or anxiety, defensive aggression and abnormal perception was observed. Abnormal perception included barking without any apparent cause, chasing light spots or shadows, and aimlessly pacing or wandering. For example, in a Belgian Malinois, a 12 nucleotide poly(a) expansion of the dopamine transporter was associated with episodic behavior modification (glazing over, lack of environmental responsiveness, episodic biting) and also with seizures. Finally, in some breeds like Bull Terriers and Cavalier King Charles, the etiology of respectively spinning/tail chasing or fly biting syndrome suggests possible links with epilepsy. In our clinical experience, compulsive or stereotypic behaviors like spinning, fly biting, freezing, shadow or light chasing were frequently associated with seizures and their frequency seemed to correspond with the severity of the attack.

In conclusion, there appears to be a clear link between behavioral changes and epilepsy in dogs. Clinicians must include epilepsy in differential diagnosis of acute behavioral modifications. Further studies are needed to determine if common pathogenic mechanisms underlie seizures and comorbid psychiatric disorders or if recurrent abnormal brain activity is responsible for behavioral troubles (epileptic psychosis).

The assessment of dog behaviour in isolation: a comparison between separation from the owner and from a cohabitant dog

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Dogs are known to form strong relationships with other dogs, and with other species. The aim of this research was to compare dog behaviour when separated from a human and a canine companion.

Sixteen dogs (9 females and 7 males, 49.8±54.3 months old, belonging to different breeds) were observed during the 2-minute isolation episode of the Ainsworth Strange Situation Test. Each dog was tested twice including one separation from its owner and one separation from a cohabitant dog. In both scenarios, 19 types of behavior were measured and compared using the Mann-Whitney test (p<0.05).

Proximity to the door (medians: 95.5 versus 54.5; Z=2.38; p=0.017), actions against the door (7.0 versus 0.0; Z=2.13; p=0.033), barking (0.0 versus 0.0; Z=2.37; p=0.017), and trying to escape from the experimental room (0.0 versus 0.0; Z=1.83; p=0.067) were higher when dogs were separated from the conspecific in comparison to separation from the owner; while passive behavior was higher when isolated from the owner (13.0 versus 0.0; Z=3.18; p=0.001).

These results suggest that dogs protest more when separated from a cohabitant dog, indicating greater intraspecific attachment. However, the heightened stress may also be due to separation from the conspecific combined with a situation where the owner was not present. It is possible that multi-household dogs spend less time alone and consequently have more difficulty in adapting to isolation, which has important consequences for dog welfare.

Heart rate vs heart rate variability: from emotions to stress responses in horses (*Equus caballus*).

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A 7 year old trained Swedish Warmblood gelding was presented for behavioural consultation due to hyper-responsiveness, increased vigilance and concentration difficulties during exercise. Firstly, a questionnaire was filled in with the owner. Secondly, using the Equine Polar System, heart rate (HR) and heart rate variability (HRV) were measured at each stage of the consultation: a baseline (B) in his own box, preparation (P) before the activity and a requested training exercise (E). In comparison with B and P, during E the horse showed increased HR, as would be expected (42 bpm vs 38 bpm vs 76 bpm), but lower LF/HF ratio scores in the HRV frequency domain results (3.0% vs 3.2% vs 2.8%). When a new exercise sequence was introduced, including an unusual stimulus, HR and LF/HF ratio jumped up to 87 bpm and 5.5% respectively.

The data suggests a control of the internal activation by the parasympathetic system during a familiar, well-controlled routine, but difficulties in coping with something new, and permanent activation before the work.

HR is useful in defining emotional activation, while HRV enhances our understanding of the stress response. Finally the LF/HF ratio score indicates differences in the autonomatic system: high values represent an unbalance in the physiological mechanisms associated with stress control. HRV is a crucial source of physiological data during consultations in Equine Clinical Ethology which enables us to accurately plan a training and re-educational programme that is adapted to the animal.

Assessment of Noise-Induced Anxiety in Dogs: Modification by a Novel Fish Hydrolysate Supplemented Diet

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The purpose of this study was to evaluate the effectiveness of a test diet (Royal Canin Canine Health Nutrition Medium Adult) supplemented with fish hydrolysate (Gabocean 3D PTP55) as a source of anxiolytic peptides, in reducing the anxiety response of Beagle dogs to noise.

The study used a parallel group design with one control and two treatment groups. Forty Beagles were initially given a baseline thunderstorm test for noise-induced anxiety. Baseline data was used in assigning control and test diets to three equivalent groups. Each group was randomly placed on a test diet supplemented with compound (0.8074% DM) or compound (0.3901 % DM) or unsupplemented control diet. Three protocols were used to assess response to the test procedure: (1) objective quantification of behavioral activity, (2) global, positive and negative anxiety observations, and (3) serum cortisol levels. The data were initially analyzed with repeated ANOVA measures. The Fisher test was used to analyze post-hoc comparisons.

The low dose test diet group showed progressive increases in inactivity over repeated testing, while the control animals performed similar to baseline; the high dose group was intermediary. The analysis of serum cortisol values revealed a significant decrease over baseline in both treatment groups for test 1 (p=0.008 low dose; p=0.0004 high dose) and test 2 (p=.001 low dose; p<.0001 high dose). By contrast, the controls showed no significant differences.

This study indicates that the fish-hydrolysate compound has anxiolytic properties in Beagle dogs, which are manifested by decreased hyperactivity and a reduced cortisol response to stress.

Interest of biological and behavioural indicators to assess the influence of pre-existing stress condition, on the emotional reactions of African grey parrots (*Psittacus erithacus*), exposed to novelty-related stress: preliminary results.

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A previous study showed the correlation between Heterophil to Lymphocyte Ratio (HLR) and stress in a population of *Psittacus erithacus*. This study was designed to compare the dynamic relationships between HLR and behavioural stress-indicators in two groups of parrots (with or without clinical signs of chronic stress), exposed to a novel environment.

A population of 33 African grey parrots, 15 females and 18 males, was divided into two groups, one (N=17) showing clinical signs of chronic stress, the other (N=16) showing no signs of stress. Every parrot was placed in a test cage for a period of five minutes; their behaviour was videotaped and droppings, if produced during the test, were collected to measure the level of fecal corticosterone (results pending). After the observation, blood smears were obtained.

Using a three-way ANOVA model (group/sex/droppings) we found that HLR showed no significant difference between the two groups (df=1; F=0.17; p = 0.6810). Using a two-way ANOVA model (sex & droppings), no significant difference was observed in HLR between males and females (df=1; F=0.48; p = 0.4934), but HLR showed a significant difference between parrots producing droppings (1.53 \pm 0.81) and those that did not (1.09 \pm 0.38) (df=1; F=4.37; p=0.0455).

These results suggest that the evolution of HLR in parrots exposed to novelty is not significantly affected by sex or pre-existing emotional condition. HLR appears to vary most in individuals experiencing generalized stress, including autonomic reactions (defecation).

The impact of the dog's behaviour profile on owner satisfaction and lifestyle

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The dog's temperament and behavioural profile have been cited as major factors that could impact the human-animal bond. The aim of the present study is to explore the influence of the main dimensions of behaviour, as measured using the CBARO¹⁰¹, on the owner's stated level of satisfaction and the perceived impact of dog ownership on owner-lifestyle. Owners of adult dogs (age >1 yo, n=785) were recruited through a UKbased dog magazine, and asked to complete an online questionnaire. This included the English version of the CBARQ¹⁰¹ scale, as well as questions on demographics, satisfaction and positive/negative impact on lifestyle. Statistical analysis was with multivariate regression and discriminant analyses (O-PLS and O-PLS-DA). An O-PLS model identified a systematic correlation between CBARQ101 factor scores and low levels of satisfaction, with "dog-directed fear and aggression", and "stranger directed aggression" providing the strongest loadings (R²X=0.253, R²Y=0.186, Q²=0.177, p=8.45 x 10⁻³⁴). Sixty people reported a negative impact of the dog's behaviour on their lifestyle, with 283 reporting no impact. In an O-PLS-DA model, CBARQ¹⁰¹ factors most strongly associated with a negative impact on owner-lifestyle included "dog directed fear and aggression", and "separation related behaviour" (R²X=0.354, $R^2Y=0.311$, $Q^2=0.243$, $p=0.66 \times 10^{-19}$). We could not produce a valid statistical model of behaviours contributing to a positive impact on lifestyle. These preliminary results suggest that the different temperament traits and behaviour problems showed by dogs could differentially contribute to the overall level of satisfaction of their owners, as well as to the perceived impact on lifestyle.

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The Cat Appeasing Pheromone (CAP): when chemical communication restore peaceful relationships between cats

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Conflict between cats in multi-cat households represents a problem for both owners and vets. However, recent research on the action of CAP has led to the synthesis of a spot-on application designed to facilitate social interaction. This study evaluates the efficacy of spot-on application of CAP in managing social conflicts in multi-cat homes.

Twelve households of aggressive feline housemates were involved in this multicentre, double blinded, placebo-controlled study conducted through five veterinary clinics. At inclusion (T0), the spot-on treatment (placebo or CAP) and a standardised behavioural programme were both administered. The spot-on treatment (placebo or CAP) was applied only one time at T0 after inclusion on each cat for all the housemates. Social relationships between cats were assessed by owners thanks to two Analogical Visual Scales ("aggression" AVS1; "affiliation" AVS2). The evaluation was performed every 10 days for 2 months (T0, T1, T2, T3, T4, T5).

Results showed that at T0 there were no significant differences between the two treatment groups regarding the two AVS (AVS1 df=9, t=0.23, p=0.82; AVS2 df=9, t=0.41, p=0.70; Student t-test). After the application of treatment, there was a statistical tendency for the AVS1 and a significant difference for the AVS2 in favour of the CAP treatment (AVS1 df=1, F=4.81, p=0.06; AVS2 df=1, F=75.63, p<0.001; mixed ANOVA model).

This research highlights the interest in feline behavioural medicine of associating a user-friendly galenic form with a maternal chemical message capable of restoring peaceful interactions between cats when conflict is already present.

Use of a double signal in a separation anxiety clinical case after failing the classic treatment.

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Canine separation anxiety is one of the most frequent behavior problems in dogs. Treatment plans such as behavior modification with medication have been established by many behaviorists. Desensitization to pre-departure routines and training to short departures are part of the classic treatment. Chloe, a 4 year old pinscher, was referred to me after the failure of the classic treatment. After being reevaluated, we decided to introduce Fluoxetine 1mg/kg SID (1) and to change the behavior management program to the double signal therapy (2). The double signal therapy consists of increasing predictability, thereby reducing stress. We also introduced the use of a babysitter when the owner had to work at night. Moreover, a safe place was created for Chloe at home. After 2 months of treatment, the owner saw a huge improvement in Chloe's behavior. Chloe didn't follow her owner all the time at home and she was more independent. After 3 months of treatment, Chloe was able to stay alone at home all night when her owner worked the night shift.

Validation of a temperament test to determine laboratory Beagles dogs adoptable as pets.

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In spite of regulatory encouragement, some laboratories are reluctant to offer their beagle dogs for adoption due to adaptation problems in some dogs.

This research was designed to encourage adoption by developing a temperament test which would select dogs that were suitable as pets. Based on a test that had been validated in a dog shelter, 6 criteria were developed in 17 subtests adapted to the laboratory context.

To validate the test, the study was divided into different stages: feasibility; inter-, intratester and test-retest reliability. 14 beagle dogs were tested twice at 3 month intervals, and a video sequence of the tests was made. Lastly, to check test predictability, 12 tested dogs were given to veterinary students for 4 weeks and each student completed a questionnaire to assess their dog's capacity for adaptation.

The test can be conducted in just 10 minutes, and can be easily carried out by technicians. The test proved reliable: Spearman's rank correlation coefficient shows a significant correlation for inter-tester (rho=0.79, p<0.001 for tester A/B; rho=0.95, p=0.001 for tester A/C; rho=0.91, p=0.02 for tester B/C), intra-tester for testers A and B (rho=0.93, p=0.04 for tester A; rho=0.64, p=0.02 for tester B) and test-retest reliability (rho=0.61, p=0.02). To conclude, the test seems to be pertinent in detecting adoptable or non adoptable dogs (predictability). However, due to a low sample size, further research is required to prove statistical correlation between test temperament score and the adoption questionnaires. Following further research, this test could encourage adoption by providing a reliable tool for the selection of suitable dogs.

Maternal behaviour in domestic dogs: a comparison between primiparous and multiparous female dogs.

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This study aimed to evaluate if maternal behaviour in domestic dogs is influenced by the number of parturitions.

Maternal care was assessed in 10 litters of different canine breeds living in a home environment. Subjects were divided into a primiparous group (5 mothers, 32.4 ± 29.3 year old; 5.0 ± 1.6 puppies per litter) and a multiparous group (5 mothers, 74.4 ± 10.0 year old, 6.8 ± 1.5 puppies per litter). Mothers and puppies were recorded by video for 15 minutes, in the morning, from day 1 to 21 after birth.

Videos of each mother and her puppies were analysed second by second using an ethogram modified from Rheingold (1963).

Data was statistically analysed using the Mann Whitney test (p<0.05) applied to days 1, 10 and 19, to compare primiparous and multiparous mothers at different stages of the post-natal period.

At day 19, compared to multiparous mothers, primiparous mothers were found to spend statistically more time in the whelping box (U=297.000; p=0.001), licking the puppies'anogenital area (U=199.500; p=0.001), in contact with puppies (U=283.000; p=0.042), and assuming postures facilitating nursing (U=253.000; p=0.012).

In primiparous mothers, on belly posture was statistically less expressed at day 1 (U=259.000; p=0.007), but more present at day 10 (U=264.000; p=0.001) and 19 (U=363.000; p=0.043).

Moreover, approaching puppies and walking within the whelping box were statistically less displayed in primiparous mothers at day 1 (respectively p=0.079 and p=0.035), 10 (p=0.015 and p=0.003) and 19 (p=0.005 and p=0.001).

These preliminary results suggest interesting differences in maternal behaviour between primiparous and multiparous female dogs.

House-soiling problem due to spongiform polioencephalomyelopathy in a cat

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A 7 month old domestic entire female cat was referred to our behavioural medicine service due to a house-soiling problem and a play-related problem described as "incompetence to play," but without aggression. The cat had never used a litter tray and eliminations were randomly distributed throughout the house. The neurological examination showed an ataxic gait and a bilateral lack of menace response. CBC and complete biochemistry panel results were within normal limits. FIV/FeLV tests were negative. Thorax X-Ray, abdominal ultrasound, MRI and CSF analysis showed no abnormalities. Since an inborn error of metabolism was suspected, the owners rejected further laboratory tests.

The owner was advised to correct the house-soiling problem by covering a relatively wide floor area with newspaper, and to use reward-based training with a clicker. No pharmacological treatment was given. After 3 months the cat began regularly using a small newspaper-covered area to defecate and urinate. The cat was later referred to the neurology service in *status epilepticus*. Neurological findings were as described before and the same tests described in the first paragraph were performed with no new results. Urine and blood analyses were also performed to rule out the most common storage diseases. All enzyme activity was within normal limits. An anticonvulsive treatment was introduced but the response was poor. After 2 years of treatment, the cat was euthanatized. Histopathology revealed a spongiform polioencephalomyelopathy. Suggested aetiologies include congenital disease (mitochondrial deficits), prion disease, and retrovirus infection. We are currently investigating the cause.

This case illustrates the importance of considering medical conditions in all behavioural cases and using an accurate diagnostic protocol.

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Case Report: The "Nutty" English Staffordshire Terrier: A Simple

Case of Hyper-Reactivity

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A 2 year-old, neutered, male English Staffordshire Terrier with a lameness problem

presented for hyperactivity, excessive nipping and jumping on people. There was

concern about the risk of inadvertent harm to children. The dog was referred by an

orthopaedic surgeon who was reluctant to operate on his elbow because of the dog's

behaviour.

The dog had been "hyper" since he was 6 weeks old, had knocked a small child over,

and nipped several young children. Many different treatment strategies (including all 4

forms of operant conditioning) had been exhausted. The dog was receiving insufficient

exercise due to the lameness issue.

A physical examination, CBC, and biochemistry panel revealed no further abnormalities

other than a left front leg lameness caused by coronoid pathology, previously diagnosed

by an orthopaedic surgeon and radiologist based on the results of a CT-scan.

The dog pulled the male owner into the consultation room, initially jumped several

times on the owner and behavioural veterinarian and repeatedly nipped and jumped up

on them throughout the consultation. The dog reacted to external stimuli, but was also

able to intermittently rest and sleep. It was difficult for the client to re-direct the dog

when reactive.

The diagnosis was hyper-reactivity.

Treatment was initiated with 1 mg/kg fluoxetine taken orally once per day. Behaviour

modification focused but was not limited to clarifying owner-animal communication.

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Risk management was explained to the clients. They were taught to use a U-Lead and, at a later date, a Gentle Leader® to improve attention training when outdoors.

The dog's behaviour improved considerably.

Feliway® Spray assessment to handle stress in cats in veterinary consultation.

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Going to the veterinary clinic is a stressful experience for most cats. This study aimed to investigate how Feliway® Spray might help reduce stress in cats and facilitate their handling, when used during a consultation.

A randomized, double-blind, placebo-controlled clinical trial was conducted using a sample of 87 cats of both sexes, aged between 7 months and 15 years with no signs of disease at the time of consultation. Feliway® spray and a placebo solution spray were tested in two consultation rooms. During the first phase, Feliway® spray was applied to the examination table in one room while the placebo spray was applied in the other. After a washout period of 15 days, the spray locations were reversed. During the first 15 minutes of general examination by the veterinarian, the stress and behavior of the cats were assessed by the observer based on a 7-level *Cat-Stress-Score* and a 5-point *Scale of Handling*. Cats exposed to Feliway® were compared to placebo cats, independent of the room location.

Results show that the use of Feliway® leads to statistically significant (p=0.003; Fisher's Exact test) differences in the behavior of cats as assessed by their owners, including a greater ease of handling compared to previous consultations. In terms of stress, animals exposed to Feliway® demonstrated significantly (p=0.021; Student's t-test) lower *Cat-Stress-Score* levels than those treated with placebo. Regarding the *Scale of Handling*, the difference in scoring was not statistically significant (p=0.1145; Student's t-test) between the two groups.

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In conclusion, results suggest Feliway® can be an efficient tool to help decrease stress in cats during consultation.

Poster presentations

Easy prescription of psychoactive drugs in dogs and cats, clinically effective in 7 days

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There are several ways to prescribe psychoactive drugs for behavioural disorders in dogs and cats: the choice of the drug is usually based on nosography, or on pharmaceutical or biochemical classifications. I propose a new prescription method, based on clinical evaluation of mood, emotions, and associated behaviours.

A simple and explicit model of emotion and mood evaluation is proposed: emotions are divided into three groups: fear-related, anger-related and sadness-related emotions; the correlated moods being anxiety, hyperarousal states, and depression. The three emotion groups are correlated respectively with the flight, fight, and freeze stress-solution strategies, and also with, respectively, action-disposition, action-avoidance and action-inhibition dispositions; associated behaviours and symptoms are linked with each category. The clinical effects of the most frequently used psychoactive drugs are correlated with emotion and mood stabilization, and with behavioural improvements.

The outcome is a simple model facilitating the prescription of psychoactive drugs that are clinically effective in nearly all behavioural disorders in a very short time.

Evaluation of the dangerousness of shelter dogs

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In recent years, the number of shelter dogs perceived as dangerous has increased. This study aims to define an objective method for assessing the dangerousness of shelter dogs (Levi et al., 2009).

41 shelter dogs (age 8 months to 10 years; 11 purebred, 30 crossbred; 14 intact females; 27 intact males) were involved in this study over a period of one year, beginning in July 2011. Dogs were housed alone in 2x4m pens. The physical (gender, size, age, mandibular thickness) and behavioural (aggressiveness towards humans or dogs) parameters of each dog were assessed by a trained examiner using an appropriate test. A second part of the test was focused on any previous episode of biting towards people or animals. Data were scored on the basis of a numerical scale (the lowest score corresponding to the lowest danger level) and analysed by chi square (SPSS 12.0, 2003).

The average test score was 30.1 ± 3.4 (range 15-95). According to the scores obtained, dogs were divided into two categories: non-dangerous (< 30), potentially dangerous (30-50) and demonstrably dangerous (> 50). 93% of adopted dogs had a score < 40. A standardized follow-up by telephone (3 months post adoption) revealed that none of the adopted dogs presented aggressive behaviours.

The use of an objective method to assess the dangerousness of shelter dogs is an important tool for veterinarians in deciding the best procedure for managing a problematic dog. Furthermore, the test proved to be a good indicator for adoptability and success in a family living situation.

Cognitive Dysfunction Syndrome progress in dogs: a 6 month-long study

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Dogs suffer from an age-related syndrome of cognitive dysfunction (CDS) that naturally reproduces key aspects of Alzheimer's Disease including $A\beta$ cortical pathology, neuronal degeneration, and learning and memory impairment. In order to develop accurate protocols for therapeutic studies and clinical procedures on both human and canine dementia, longitudinal studies are needed.

The aim of the present study was to assess the advancement of canine CDS during a sixmonth period by using an owner-based observational questionnaire (González et al., 2011).

Fifty-two elderly dogs (≥9y.) were enrolled in the study and sorted into three groups: i) Cognitively Unimpaired (CU), n=25, ii), mildly Cognitively Impaired (mCI), n=16; and iii), and severely Cognitively Impaired (sCI), n=11.

A Wilcoxon test was performed to compare questionnaire results at baseline and after six months.

Only mCI animals showed significant differences across the period of study with a higher total dysfunction score (basal score of 7.75±0.86 versus a score at follow up of 9.19±2.51; p<0.05) and a trend towards an increase in the number of affected items (p=0.06) at the end of the 6-month period. Questionnaire results after 6 months did not change from baseline in CU and sCI animals.

Considering the present results, the observed advancement of the disease in mCI dogs might stress the need for clinical consultations in these animals more than twice a year

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in order to establish appropriate therapeutic measures. However, a longer period appears necessary to detect behavioral decline in unimpaired or severely impaired dogs.

Cat and Dog Behavioural Problems in Portugal

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Data about national veterinary practice realities is essential for professionals dealing with behavioral problems, not only for managing cases but especially for prevention. This study aims to shed light on Portuguese veterinarians' attitude to behavioral problems, and to estimate the most frequent complaints and methods used to treat these in dogs and cats. A questionnaire was sent to veterinarians in 2010 to obtain information about behavioral problems and the strategies used to address them.

Of all questionnaires, 272 were valid and analyzed. Veterinarians that had been consulted for behavioral issues (n=251; 93%) reported the frequency of these visits at less than 20 times/year (dogs: n=190; 75.4%; cats: n=214; 85.3%). However, some stated that "Behavior isn't a veterinary concern" (n=7; 2.5%). The majority (n=168, 62.5%) referred some cases to a specialist, which represented less than 25% of the cases (n=132; 75%). Some (n=10; 3.6%) tried to solve all the problems for which they were consulted.

Cats: the most frequent complaint was "House Soiling" (n=193; 25.8%), followed by "Aggression to People" (n=155; 20.7%) and "Furniture Scratching" (n=127; 17%). "Neutering" (n=233; 31.6%), "Environmental Enrichment" (n=193; 26.2%) and "Behavioral Modification" (n=111; 15.1%) were the most common treatments.

Dogs: the most common complaint was "Aggression to People" (n=135; 19.2%), followed by "Destructiveness" (n=131; 18.6%) and "Aggression to other Dogs" (n=120; 17%). The most common treatments were "Behavioral Modification" (n=244; 33.1%), "Neutering" (n=190; 25.7%) and "Environmental Enrichment" (n= 138; 18.7%).

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The fact that some practitioners never referred any cases and that others rejected behavior as a veterinary issue probably reflects a gap in curricula on this topic. Improving veterinary education in this area would probably help in the prevention and management of behavioral problems.

Play agression or normal behavior between cat and dog?

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Canine aggression is one of the most serious behavioral problems; it is also one of the problems that owners and behaviorists are most aware of. In this case the owner described a problem between Peluso, her cat (a 6 month old mixed Persian) and Jack, her dog (a 3 year old Andalusian mouse-hunting dog), who showed a lack of self-control while playing. A treatment was proposed including a behavior management program, fluoxetine (1mg/kg SID) (1, 2,), pheromonotherapy and environmental enrichment for both Peluso and Jack. After two weeks of desensitization and counterconditioning treatment, Jack could stay still in the presence of Peluso. He could also tolerate the presence of Peluso at a distance of 2 meters.

At present, after creating private areas which they perceive as safe places, they accept each other's presence. It is important to understand the complexity of reactions when mixing different species. Animals communicate with each other differently depending on the species and coexistence may not always be adequate. This should be taken into account when adopting different species in the same household.

Castration in dogs behavioural problems: is it really ineffective?

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Castration in dogs decreases gonadal hormone levels which might decrease sexually dimorphic behaviours, including sexual arousal, masturbation, urine marking and perhaps some forms of aggression. However the results of neutering studies have been somewhat conflicted.

The aim of present study was to discuss the role of castration in behavioural medicine by presenting five clinical cases.

Cases 1, 2, 3 and 4 were presented for behavioural consultation. Based on the diagnosis, a behavioural and/or drug therapy was applied. Castration was later included to support and/or improve the therapeutic approach after careful evaluation of the case. Dog 5 was inadequately castrated prior to behavioural consultation by the practitioner. Follow-ups were conducted for all dogs until the conclusion of the case. For each case diagnosis, therapy and outcome are discussed.

- 1. Beagle, 3 years, displaying protective, territorial and predatory aggression, excessive sexual behaviour. Symptoms decreased in two months.
- 2. Border Collie, 3 years, displaying intermale aggression. Treated with cyproterone acetate and carbamazepine. Improved in 4 months.
- 3. *Cane corso*, 2 years, displaying aggressive reactions in prevention of resources and interdog social behaviours. Friendship behaviours reappeared in one month.
- 4. Mixbreed, 2 years, monocryptorchid, displaying aggression over resources, territorial aggression, interdog aggression, aggression towards unfamiliar people. Improved in 4 months.
- 5. Mixbreed, 3 years, displaying hyperactivity/hypersensitivity syndrome with impulsive aggression towards unfamiliar people. Treated with drug therapy (fluoxetine), training programme. Improved in 4 months.

These findings partially disagree with previous data in the literature. Castration can effectively have a positive role in controlling dog behaviour when appropriately included in a global treatment rather than as a primary means of intervention.

Saturday, November 22nd 2014

SEMIOCHEMISTRY AND MANAGEMENT OF ANIMAL POPULATIONS.

2nd IRSEA International Congress

Oral presentations

Using semiochemicals in the management of wild animals: myth or promising reality?

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Human have always tried to control the other animals, to protect themselves against predators or challenging species, but also to get some direct benefit. Over millenaries, such strategies consisted mostly in hunting, poisoning, trapping or enclosing the animals. Recently, our specie became aware about the price to pay: destruction of wildlife and genetic losses, damaged and unbalanced ecosystems, development of resistancy against pesticides and antibiotics, toxic side-effects affecting valued species and humans. This evolution of mentalities and understanding, has led to try to develop more sophisticated and respectful strategies, which, very often, fail in solving the problems. The purpose of this paper is to review some literature about the use of semiochemicals in the control of animals, but will also describe new possibilities, resulting from the better understanding of chemical communication. The use of pheromones has been a source of great expectations in the management of crops destructing insects. Most between those strategies were very naive, and were seen as restricted to invertebrate species. More recently, the development of pheromonatherapy in domestic species, and of semiochemical strategies as well (including the interspecific communication), have enriched our possibilities, and opened new doors to a world where humans and animals may live together, sharing the resources.

Evidence of a semiochemical as inhibitor of infesting behavior of Lepeophtheirus salmonis copepodits in Atlantic salmon smolts

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Lepeophtheirus salmonis is a copepod ectoparasite responsible for several types of damage in salmon production. The identification of putative copepodit attachment inhibitors was accomplished by comparing the cutaneous mucus obtained from Atlantic salmon and Rainbow trout in infested and non-infested farms. Preliminary investigations have identified a semiochemical that limits the attachment of copepodits. The purpose of this paper was to present the validation of the Sealice Copepodits Attachment Inhibiting Semiochemical (SCAIS) during an infestation test, using an experimental (SCAIS) group, a positive reference group (non-treated salmon) and a negative group (cod).

Naive salmon were treated with a 6ppm SCAIS solution, for 10 minutes, and then bathed in a 3 ppm solution for the duration of the entire test. The reference groups (salmon/cod) were treated with the solvent following the same protocol. During the test, all fish were exposed to 60 copepodits for 45 minutes, and then euthanized. The entire surface of their bodies were scrubbed to obtain the mucus; this material was then filtered and the copepodits counted. Data were analyzed using a one-way ANOVA and Tukey's test.

The reference salmon (n=14) were infested with 20.8 ± 3.4 copepodits on the body. The SCAIS salmon (n=16) showed significantly lower infestation on the body with 6.3 ± 2.6 copepodits (p<0.001), and the cod (n=7) had 4.7 ± 2.3 copepodits (p<0.001). There was no significant difference between SCAIS salmon and cod (p=0.47).

This semiochemical appears to inhibit the stable attachment of copepodits on salmon, leading to a copepodit infestation comparable to that of a non-host fish species. Further

study should assess if the copepodits attached in such contexts remain attached and continue the development cycle.

Interest of a semiochemical to protect horses against Simulid ear flies

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Horses are commonly harassed by various species of haematophagous arthropods, whose bites constitute a major cause of stress. One of these insects, Blackflies (Diptera, Simulidae), bites horses inside their ears and generates acute stress, lesions and allergic reactions. Despite various chemical products and protection material, no real effective protection is currently available. The aim of this study was to assess the efficacy of a semiochemical, derived from a Mustelid/Viverrid secretion, to prevent the infestation of the horse ears by this diptera.

A randomized and blinded paired-samples study was carried out on 16 horses. For each horse, insects present in the ear were removed, the inside of the ear was washed, and a treatment applied. Each horse received either a placebo or the semiochemical treatment on its ears (randomization stratified especially concerning laterality). One hour after the application of treatments, 2 independent observers counted the number of simuliidae present in each horse's ears.

A median number of 1.25 insects were found in the placebo ear compared with 0 insects in the treated ear. A significant difference between treatments was observed (Z=3.30; p=0.001, Wilcoxon for paired-samples test). The tested semiochemical provided 90% protection against the infestation of horses' ears by simuliidae over the course of 1 hour.

Additional studies need to be performed to further investigate the duration of efficacy for this semiochemical. Preliminary data highlights the potential interest of the semiochemical approach as a new strategy for fighting hematophagous insects. Such treatment could be used to improve horse welfare without the use of toxic or banned xenobiotic substances.

Predator-prey relationships-related semiochemicals between terrestrial gastropods ans predacious ground beetles species as tools for investigating new pest control means

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Predation has long been recognized as a major selective force that drives the evolution of organisms, shaping their life history traits, morphology, ecology and behavior. Cues that allow prey to avoid predators without coming into direct contact appear particularly suitable to improve chances of survival. In this regard, scientists have recognized the efficacy of chemical cues which betray the presence of predators, and these cues have been well-documented in numerous taxa. However, there is limited data available on terrestrial gastropods, whose main activities and biological interactions are chemically mediated.

Over a series of trial choice experiments, we demonstrated that putative cuticular compounds from predatory ground beetle species significantly altered the self-maintenance behaviors of terrestrial mollusks. In the first experiment, the pest slug *Deroceras reticulatum* spent significantly more time in the control shelter than in the shelter treated either with *Carabus nemoralis* or *Carabus coriaceus* cuticular extracts (Z=2.43; p=0.0151 and Z=3.31; p<0.01 respectively, Wilcoxon matched-pairs signed-ranks test). In the second experiment, cues from *Carabus nemoralis* significantly impaired food intake of *D. reticulatum* compared to a control (Z=2.19; p= 0.028 Wilcoxon matched-pairs signed-ranks test). In another two-choice test, we assessed the climbing reflex response of an invasive xerophilous alien snail species, *Xeropicta derbentina*, in the presence of different predacious ground beetle body extracts. Only *C. morbillosus* significantly altered the climbing area choice of the snail (p=0.019; Student t-test) but the results confirm some aspects of previous findings obtained with *D. reticulatum* and are supported by eco-evolutionary concepts.

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What can we expect from semiochemicals in the management of canine behaviors?

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Introduction

Behavioral disorders are a major reason for the breakdown in the human-animal bond and can result in a dog losing their home. Behavioral disorders range from normal but undesirable behaviors, to dangerous behaviors and behaviors that are abnormal. The goal of the veterinary practitioner is to help the pet owner with the undesirable behaviors and restore the human-animal bond.

The treatment of canine behavioral disorders utilizes many different modalities. Although the goal of all of them is to change undesirable or problematic behaviors, some treatments are more human centered than dog centered. Behavioral modification requires the pet owner to understand how to apply the techniques and make time to work with the dog often difficult for owners to accomplish. Psychotropic medications require an accurate diagnosis and the understanding of pharmacology and owner compliance to make sure the pet is given the medication daily. Semiochemical intervention (pheromonatherapy) is unique in that once they are applied they are easily recognized and responded to by the dog and requires minimal human intervention to be effective.

Why Semiochemicals?

Although dogs communicate in many ways, visual, tactile and auditory, semiochemicals reach the dog through one of their most sensitive modalities-olfaction but more specifically through the vomeronasal organ (VNO) and onto the accessory olfactory bulb in the brain for processing. Semiochemicals are used to identify species, individuals, group members and also perhaps gender, age and health. When using semiochemicals therapeutically, we often use concentrations stronger than those found naturally to help make the signal more salient and increase the animal's sense of security. However, effective use of pheromonatherapy relies on knowing both when they are appropriate and effective and when they may not be useful.

What are pheromones and what is pheromonatherapy?

The definition of pheromones may vary so for the purpose of this paper and presentation the following definition will be utilized

"In animals with a limbic brain, the term 'pheromone' refers to chemical signals which are normally used in intraspecific communication, which are typically detected through the VNO and which appear to have an intrinsic effect on the emotional processing of the receiver."

With this definition we can include alarm pheromones, sexual pheromones and other types of pheromones that may influence behavior.

In dogs olfactory substances play a large role in communication with the pheromones in the area of the face, mouth, anal area utilized in recognition and fear based pheromones may send signals to other animals and certainly can increase stress and anxiety in animals nearby. Appearing pheromones help the formation of the maternal bond between a bitch and her pups.

Pheromonatherapy is "the use of chemical signals that are involved in intraspecies communication in the clinical setting to manage behavior".

Pheromones are not "aromas" and not the same as aromatherapy. Aroma therapy is based on plant based mixtures, while pheromones are based on animal signaling processes. Pheromones are an unconditional signal that brings about change without the necessity of prior learning.

The primary semiochemical in canine pheromonatherapy and its use

The pheromone utilized for canine behavioral therapy is an appeasine (known as dog appeasing pheromone), which occurs naturally in dogs around the mammary area of a nursing female, and a similar compound can be found around the ear of an adult dog. The synthetic copy is marketed as Adaptil®¹ which is available as a spray, diffuser and impregnated collar. Pheromones affect behavior within a certain context and Adaptil is most effective on the emotional responses of pain, anxiety-fear, panic, frustration and grief. They are best utilized in situations dealing with physical and social stimuli. However several considerations are important when contemplating the use of pheromoatherapy.

 An appropriate diagnosis of the problem and understanding of the underlying emotional responses causing the undesired behavior. If frustration is a major motivation for the behavioral response, pheromones may not be as effective.

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- Pheromones do not replace pharmacotherapy, but can be synergistic and it may be necessary in some situations to use both.
- Placement of diffusers is critical to achieve best access to the responses.
 Diffusers should not be blocked by furniture, near air ducts or open windows.
- Individual variation in responses to any behavioral treatment intervention
- Risk for the handler is low as is risk for the patient. While no intervention is without risk toxicology studies conducted to date have not shown problematic issues.
- Pheromones work by encouraging expression of certain behaviors but not by directly creating or inhibiting certain behaviors.

Behavior disorders susceptible to pheromonatherapy intervention in the dog

Several behavioral disorders in dogs respond to pheromonatherapy either alone or in combination with other treatment modalities and these responses are often supported by research trials.

Separation related problems

Separation related problems are quite common in dogs with cases making up 17-40% of the referred caseload. The most commonly cited signs include destruction, vocalization and elimination, which occur when the dog is home alone. However, often the problems are lumped together as separation anxiety when in reality it can have different underlying motivations. A dog may be agitated and anxious by being separated from individuals to whom they are strongly attached, or anxious because something bad has happened to them when home alone and are afraid to alone again, have a generalized anxiety disorder that worsens when alone, and finally a dog may not be anxious at all but frustrated by outside influences, limited enrichment and opportunities to express normal behavior. The accepted behavior modification treatment protocols include independence training, uncoupling departure cues from departure, making departures less stressful (treats, activities), creating a safe haven, creating predictable interactions and training departures. Pheromones have shown efficacy in the treatment of separation related problems in dogs with the best results shown in destructiveness and vocalization behaviors when compared with Clomipramine. These treatments are often accompanied by medication, two of which have approval for use in dogs; Reconcile and Clomicalm. The use of pheromones is especially important due to the rapidity with which they work allowing the dog to calm as other treatment modalities are applied. It is a well-accepted intervention in nearly all cases of separation distress syndrome to add in pheromonatherapy.

Sound sensitivity

Many dogs are quite sensitive to loud noises including fireworks and thunderstorms. In a retrospective study of diagnoses at a university behavioral referral center, noise phobias and separation anxiety occurred together more often than chance would predict therefore when dealing with one or the other it is prudent to screen for them both. However, it is likely that sound sensitivities go unreported when the signs are not severe or do not interfere with the quality of life for the dog or owner. The most frequently noted signs include panting, trembling, restlessness and hiding. Less frequent signs such as destructiveness and elimination are reported.

Several studies have shown the effectiveness of pheromone therapy for the treatment of firework phobias in dogs either in conjunction with medication, CD training program or alone. For best results the diffuser should be in place at least a week before the event. It is also advisable to create a safe place for the dog and train them not only to go there, but also associate the location with good things like food, treats and quiet attention or massage. Pheromones will not sedate the animal, in fact they may still startle at loud noises but more quickly return to baseline because it may process the event in a less anxious manner. Pheromones seem to work best in conjunction with a desensitization program. Results with pheromones and a behavior modification program often seem to be carried over from one event to another with lasting behavioral changes.

Adaption to a new home

Moving to a new home can be stressful for dogs and cats. Creating an environment that is as familiar and non-threatening as possible is desired. During the packing and transition, use of a pheromone-impregnated collar can help calm a dog. Using Pheromones pre-emptively with installed diffusers before the dog enters the environment may help the dog adjust more quickly to their new surroundings. Pheromones are also indicated when bringing a new dog home from a kennel or shelter.

Travel related problems

Many dogs show a wide range of unwanted behaviors when traveling in the car. These range from vocalizations, jumping, running around, salivating, vomiting and cowering. Travel related problems can be due to excitement (running, barking, jumping, restlessness and failure to follow commands), anxiety-fear (remaining close to the

owner, trembling, licking, panting and startling) or even nausea (vomiting, salivation, hiding, cowering) but often it is difficult to know all-underlying motivations and the categories may not be mutually exclusive. Travel related problems are not often referred to specialists but can be a reason for limited visits to the veterinarian or for the dog to experience other outdoor experiences that would be enriching. A study by Gandia Estella and Mills indicated the use of a pheromone-impregnated collar was effective at managing and improving travel related problems by a decline in the signs and with limited relapses after 6 weeks of therapy. For fear and anxiety related signs improvement was not always sustained when pheromone treatment was discontinued perhaps necessitating longer treatment times.

Puppy behavioral development

Pheromonatherapy is useful when bringing a puppy to a new home and the use of a diffuser and an impregnated collar is recommended at that time. Puppies that were exposed to dog appeasing pheromone may show a reduced rate of vocalization over night in the first few weeks when compared to puppies with a placebo diffuser or collar. Many studies have investigated and verified the effect of attending puppy class to aid in the appropriate social development when acquiring a new puppy. Pheromonatherapy has been shown to particularly useful for puppies between 10-15 weeks who wore pheromone-impregnated collars (Adaptil) for the 8 weeks of puppy classes when compared with puppies wearing a placebo collar. Data from telephone follow up of both groups showed a statistical difference in socialization and fear-based behaviors between the treated group and the control group with results noted not only during class but also up to one year later. Puppies who wore the collar did better in puppy class and were more sociable at one year of age.

Adaptation to the veterinary hospital

Veterinary practitioners are aware that many dogs are very fearful and anxious when arriving to the veterinary hospital and during routine procedures. Some of these dogs may also suffer travel anxiety that carries over into the clinic and can be addressed with pheromonatherapy. However, some dogs are fine in the car, but very anxious once at the clinic. One study indicated that Dog Appeasing Pheromone in the veterinary clinic (waiting area and examination room) reduces signs of anxiety but not aggressive responses in dogs. Because this is such an easy intervention, the use of diffusers in

waiting areas and examination rooms is recommended along with low stress handling and fear free examination techniques to decrease anxiety, stress and fear in dogs.

Dog appeasing pheromone has also been shown to decrease acute phase perioperative stress in dogs. Forty six dogs housed in animal shelters underwent elective orchiectomy or ovariohysterectomy and were either exposed to sDAP or sham treated with just the carrier portion 20 minutes prior to use and put into the cages 30 minutes prior to surgery. Dogs exposed to sDAP were more likely to be alert and exploratory after surgery than dogs exposed to the sham treatment.

Adaptation to the kennel environment

Dogs in kennel situations whether in the veterinary clinic or shelters can undergo stress and anxiety. Improving the welfare of kenneled dogs is extremely important and many articles address these situations in various ways including enrichment, music, and feeding enrichment toys. Semiochemicals have also been utilized with Dog appeasing Pheromone reducing barking amplitude and frequency in dogs exposed to diffusers for 7 days. There were also significant differences in resting, barking, sniffing and responses to strangers. If diffusers are not a practical option in the kennel, Dog Appeasing Pheromone in a spray form can be applied to bedding before introducing the dog to the area. In other situations if the kennel allows, pheromone impregnated collars may be appropriate.

Conclusion

Semiochemicals in the form of pheromones are extremely useful adjunct to behavioral therapy in dogs and can help reduce anxiety, fear, distress associated with various behavioral conditions. Not only are they easily recognized by dogs, but are also easily administered by owners aiding in compliance with treatment plans.

When rodents arrive to avoid predators: the role of olfaction?

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Following humans, rodents are the most widespread family of mammals in the world and are found everywhere from desert climates to subarctic regions. Their size, wide distribution, and number lead to a vast array of predators, such as kites, owls, falcons, kestrels, harriers, corvids, cats, foxes, snakes, ferrets, weasels, stoats, mongooses and dogs.

The study of anti-predator behaviours in rodents has attracted the interest of numerous studies, from psychiatric models of stress conditions, to biochemical and genetic studies linking the implicated brain areas, genes and receptors for predator molecules, and more applied studies regarding the control of populations.

In laboratory animals, different models of samples and animals have been used to obtain a predatory response under controlled conditions. These experiments relied largely on the use of cats, so much so that they came to be called "the cat odours paradigm". The samples used included urine, fur, medical gauzes rubbed over the cat or their marking areas, and collars. In some of the studies, live animals were used directly. Secondly we highlight a fox odour molecule, TMT (trimetile-thiazoline), with a more controversial effect regarding the fear reaction; the latest publications indicate an unpleasant odour, and the stimulation of the trigeminal nerve. Other species tested include dogs, rats (for mice), ferrets, snakes, stoats and large felines such as lions.

Although rat and mice anti-predator responses under laboratory conditions are the most well-documented in the animal kingdom, we lack knowledge of some of the principal semiochemical compounds which mediate this response, along with real field applications.

Chemical signaling in rodents in view of rodents management.

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Rodents depend on chemical signals for survival because of their secret mode of life. These chemicals signals are excreted in urine, feces, vaginal fluid, and scent glands. It is believed that these chemical signals can be used for developing eco friendly kit to manage rodent population. The study was aimed to identify the pheromonal compound in laboratory rat (*Rattus norvegicus*), house rat (*Rattus rattus*) and field rat (*Millardia meldata*). In addition, the volatile chemical signals are believed to be transported through carrier protein, specifically the lipocalins. So the present study also dealt with proteomic analysis in the pheromone sources (urine and scent glands). The results indicate that certain volatile compounds were species and sex specific. Further, alpha 2U-globulin (18-20 kDa), a specific lipocalin was characterized at molecular level. It was found to be present in all the three rat species analysed. Based on the above findings, it is postulated to develop a pheromone trap by using sex and species specific chemo-signals along with alpha2U-globulin protein. The kit will be environment friendly because both these pheromones and carrier protein are naturally produced and can be degraded safely.

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Interest of semiochemicals in the management of inappropriate behaviours in livestock species

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The objective of this paper is to review some of the evidence showing that the use of semiochemicals –and particularly pheromones- can be useful to improve the welfare and performance of farm animals by modifying their behaviour or by reducing their stress and fear responses. Pheromones are a particular class of semiochemicals which play an important role in intra-specific communication in a wide variety of animals, including livestock species. One example of pheromones that are particularly useful in farm species are the so-called appeasing pheromones. Appeasing pheromones have been identified in several species and are produced by lactating females after giving birth. Appeasing pheromones act upon the offspring reducing aggression and fear; interestingly, adult animals also respond to appeasing pheromones when exposed to them. The pig appeasing pheromone (PAP) has been shown to be effective in reducing aggressive behaviour after mixing of unacquainted pigs. PAP is therefore useful in weaning and growing pigs when subjected to social mixing, and also in group-housed pregnant sows kept in dynamic groups. Tail-biting is a serious welfare and economic problem in growing pigs that has a multifactorial origin. Although the underlying mechanism of tail-biting is not completely understood and further research is needed on the potential benefits of using PAP to prevent it, the purported link between some types of tail-biting and aggression would suggest that PAP may indeed be useful to prevent some cases of tail-biting. This paper will also consider the potential benefits of using other pheromones in farm mammals and poultry.

First description of vomeronasal organ inflammatory changes in pigs

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The vomeronasal organ (VNO) is implicated in pheromone detection and thus in the control of animal behaviour. Thus far, VNO inflammatory changes have been briefly described only in cats, and it is supposed that they could influence behavioural problems stemming from an organic cause. The aim of this study was to investigate, using histological and histochemical methods, if the porcine VNO can be affected by inflammatory processes.

Thirty-five VNOs from 18 six-month-old female pigs were sampled and submitted to routine tissue processing. Four-µm thick sections were stained with Haematoxylin-Eosin, PAS and Alcian Blue. Inflammatory populations, necrosis and vascular congestion were evaluated in the respiratory and sensorial portions of the VNO.

All of the VNOs presented inflammatory changes affecting the respiratory epithelium with weak or moderate intensity. The sensorial epithelium was weakly or moderately altered in 23/35 (66%) of the VNOs. All these alterations were chronic in nature, and the cell population was mainly composed of lymphocytes, followed by macrophages, plasma cells, neutrophils and eosinophils. VNO epithelium necrosis and congestive phenomena were also noted. In 28/35 (80%) of the VNOs, the presence of lymphatic follicles was observed. Analysis of PAS and Alcian Blue stains showed that the mucous glands were mainly located in the respiratory part.

Our analyses demonstrated that the porcine VNO can be affected by inflammatory changes. The respiratory portion was more commonly altered than its sensorial counterpart, suggesting that it plays a more important role in VNO immunity defence. These preliminary data provide the rationale to further investigate the implication of VNO lesions in the onset of behavioural problems.

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Ovulatory signals as biomarkers for estrus: A diagnostic tool for successful artificial insemination

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The release of ovum from ovary to the fallopian-tube is known as ovulation. The timing of ovulation along with the coordination of the sexual behavior is very important to insure successful fertilization. The female mammals try to advertise the process of ovulation through prospective and receptive sexual behavior along with the morphological changes. The morphological changes include sexual skin swelling, changes in colorization genitals etc. Most importantly, animals release specific chemical signals through their scent sources during estrus to attract the male for matting. These ovulatory signals play a significant role in all the mammalian species with the exception of primates. In the current study, we report ovulatory specific signals in rodents and farm animals. The estrus specific signals were correlated with the change in the hormone profile during estrous cycle. A clear transition of the volatile compounds was recorded in the urine of rodent and farm animals during estrous cycle. Even though these compounds are excreted as metabolites of estrous cycle, these metabolites are used as chemical signals by the conspecifics to identify the reproductive status. These estrus-specific chemo-signals can be used for trapping the rats and improving the reproductive performance in farm animals. Hence, these pheromone based techniques can be used for rodent pest management and improvement of livestock production.

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Volatile identification in male Kangayam breed cattle, Bos indicus

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Background: Kangayam breed cattle (Bos indicus) are one of the indigenous livestock species known for the superior drought qualities and adaption to poor nutrition and longevity. Due to various factors this breed had started dwindling and faces extinction for the past two decades. Hypothesis: Most of the mammals depend on chemical signals for their communication, so it is possible to use these signals to rectify the hurdles in the animal communication. Pheromones are air-borne chemical substances released through urine, feces, vaginal fluid and scent gland of animals and elicit both behavioural and endocrine responses in conspecifics. Hence in the present study it is proposed to develop a novel technique by means of pheromones to improve kangayam population. Methods: Urine samples of male Kangayam breed cattle (prepubertal, heifer, intact, and castrated) were analysed for volatile compounds through GC-MS. Results: The GC-MS analysis of urine revealed a total of 36 compounds, among which the prepubertal and heifer contain eleven specific compounds, whereas intact and castrated male contained seven volatile compounds. The significant finding is the presence of male specific compound, 1-iodo-2-methyl undecane which was present in heifer, normal and prepubertal and absent in castrated animal. Conclusion: The pattern of volatile compounds identified was seems to be depends on age and physiological status of individual animal.

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Influence of Semiochemical and Odorous information on Human Behavior

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In most species, there are two chemosensory systems, both located in the nasal cavity but physiologically and anatomically distinct.

The main olfactory epithelium is principally involved in the airborne odor perception, whereas the vomeronasal organ in the detection of pheromones that are chemical compounds secreted or excreted by individuals of the same species.

Pheromones are secreted through sweat or urine and perceived by another individual that evoke a behavioral or neuroendocrine response in particular on sexual behavior. A group of pheromones secreted in the breast region, immediately after childbirth, seem to have satisfying and soothing properties of both young and adult animals that have been defined 'appeasiness' or 'appeasing pheromones'.

The HAP seems to be effective on selected human behaviors.

Will be described preliminary results of a study in which patients with different psychiatric conditions, exposed to HAP, report changes in their psychopathological condition.

What's up in mammalian semiochemical's chemoreception? An update on the pheromone-responding receptors.

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Chemical communication is widely used among living beings to exchange information. In mammals, the chemoreception of semiochemicals is carried out by at least two major sensory systems contained in the nasal cavity: the vomeronal organ (VNO) and the main olfactory epithelium (MOE) which, contrary to previous beliefs, have an overlapping role in pheromone detection. Since Dulac and Axel's work in 1995, researchers have continued to discover chemoreceptors in both systems, which are capable of recognizing a wide variety of chemical signals and structures that produce known pheromonal effects. The aim of this review is to describe these new findings in chemoreception.

In mammals' VNOs, semiochemicals can be detected by three types of G protein-coupled receptors: (i) the V1R, expressed in neurons of the sensory epithelium and activated by volatile ligands; (ii) the V2R, found only in some mammalian species such as rodents, and activated by high molecular weight, non-volatile pheromones such as peptides or proteins; (iii) the Formyl-Peptide Receptors (FPRs), a novel class of chemoreceptors whose ligands are microbial and viral peptides. These receptors are associated with a different α -subunit of G protein to transduce the signal and are involved in different signalization cascades.

The MOE in mammals is also capable of detecting pheromones via neurons expressing V1R or another family of receptors named Trace Amine-Associated Receptors (TAARs) specialized in the detection of alarm pheromones. The last sensory system involved in chemoreception has been recently rediscovered: the Grueneberg Ganglion, where cells expressing one V2R type and TAARs have been reported.

Poster presentations

Toward a new fight method by host imposture against the brown dog tick?

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Rhipicephalus Sanguineus is the most widespread tick in the world and is capable of transmitting a great variety of infectious diseases. Because of the proximity and relationships between men and dogs, Rhipicephalus sanguineus is a major public health issue. Faced with the resistance of ticks to acaricides, developing new ways to fight them has become critical, and the use of chemical communication could be a solution. The aim of this study was to assess the effect of non-attacked host secretion compounds on the host-seeking behaviour of Rhipicephalus Sanguineus.

To increase the tick's motivation to crawl while displaying host-seeking behaviour, a host-associated attractant stimulus system was developed (the "attractive cylinder"). Four treatments were tested with this system: three solvent solutions with the active product in different concentrations: 5% (i.e. 0.2μl/cm²), 2.5% (i.e. 0.1μl/cm²), 1.25% (i.e. 0.05μl/cm²) and a control (pure solvent). 20 ticks were used per treatment. Each tick was placed alone on the device for a maximum of 2 minutes. The number of ticks that performed climbing behaviour on the cylinder was noted.

Results showed significant differences between control and the different treatments (chi-square test): 5% (df=1 χ^2 =14.4 p=0.0001), 2.5% (df=1 χ^2 =6.67 p=0.0098), and 1.25% (df=1 χ^2 =5.23 p=0.0222) regarding the number of ticks climbing on the cylinder.

These first results indicate the potential of a new method for controlling ticks: creating a disturbance in the host perception using a semiochemical approach. Further research should explore in greater detail the possibility of using applied semiochemistry.

Innovative approach on semiochemistry: The electrical penetration graph (EPG) applied to Hemipterans insects

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Insects rely on many different senses (i.e., vision, chemoreception, mechanoreception, hearing) to achieve a wide array of specific behaviors. Because of the diversity of these sense mechanisms, insects are often not well understood, and many aspects still remain unexplored.

The invaluable assistance of electrophysiology techniques developed over the last decades has significantly improved knowledge by unraveling the underlying sense mechanisms. This is particularly true regarding the study of the cryptic feeding behavior of piercing-sucking insects, the understanding of which has seen incredible advances with the advent, in the 60s, of the "Electronic Measuring/Monitoring System" or EMS.

The device has been improved and is now known as EPG. It has been intensively employed in studying feeding in aphids and others hemipterans, and to investigate the transmission mechanisms of associated diseases into host plants.

We aimed to characterize the main feeding stages in two hemipteran pest species with slightly different feeding modes: *Hyalesthes obsoletus*, a potent homopteran vector of a devastating disease affecting lavender found in southeast France, and the Sycamore lace bug *Corythucha ciliata*, a heteropteran alien species.

Using a combination of semiochemistry and EPG technology, we will be able to test different substances in order to change the feeding behavior of insects. The goal of future studies will be to find ways to keep insects from biting and to deflect food intake that occurs in the phloem in order to prevent disease transmission.