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A TAXONOMY BASED EVALUATION OF *IN VITRO* CELL-MEDIATED IMMUNE EFFECTS OF ALCOHOLIC PLANT EXTRACTS IN PIGS FROM A LOW-INPUT FARM

Diana Olah¹, Carmen Dana Șandru^{1,3}, Marina Spînu^{1,3}, Mihai-Horia Băieș¹, Aurel Vasiu¹, Laurian Vlase² Ana-Maria Vlase², Gianina Crișan², Vasile Cozma¹

¹Department of Clinical Sciences, Division of Infectious Diseases, University of Agricultural Sciences and Veterinary Medicine, Faculty of Veterinary Medicine, Cluj-Napoca, Romania

²Department of Hygiene and Bromatology, Faculty of Pharmacy, University Iuliu Hatieganu, Cluj

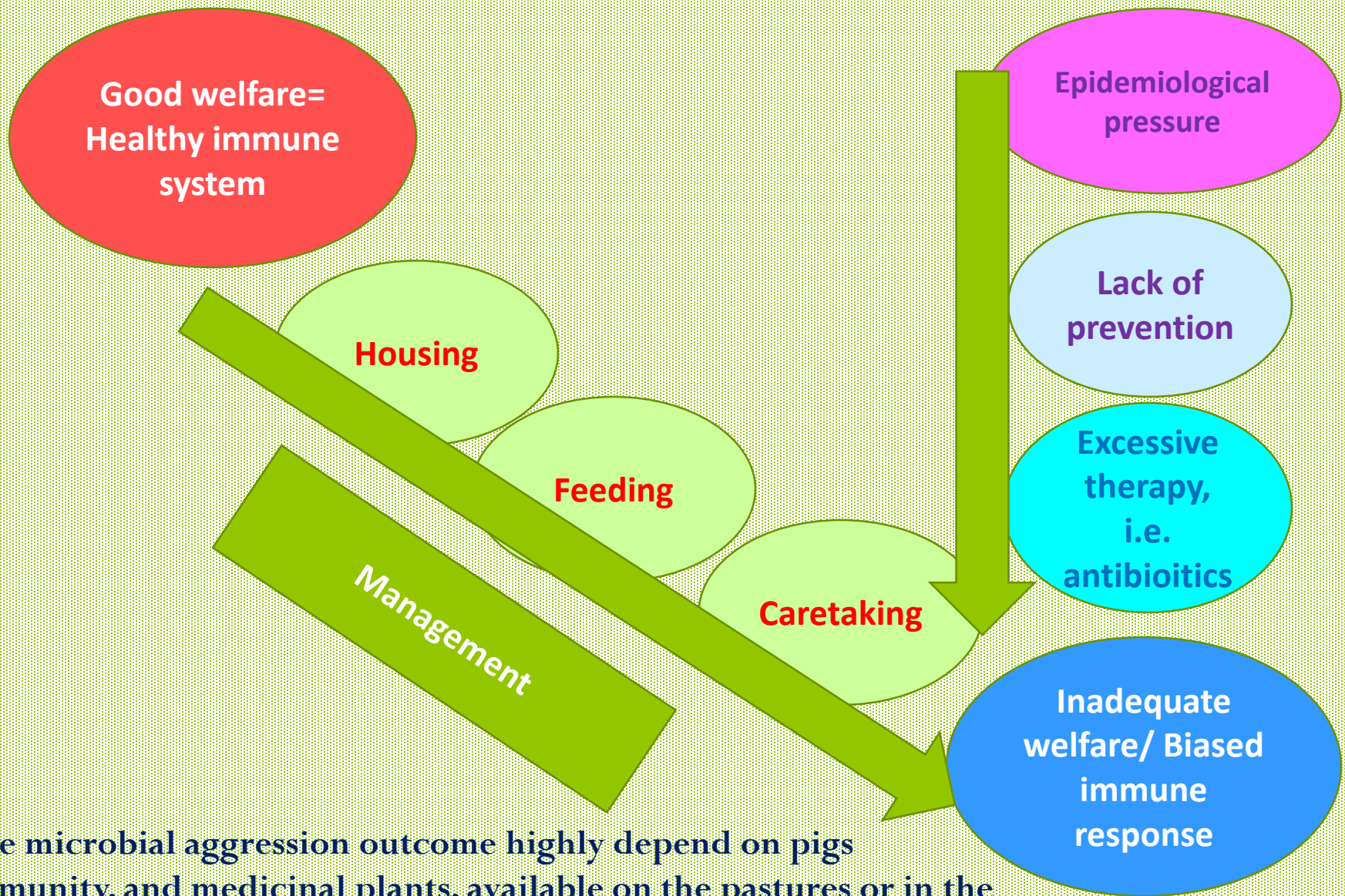
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Poultry and Pig Low-input and Organic production systems' Welfare

- In Romania, swine breeding is an important tradition, the vast majority of pigs being raised in low input systems. Recently an increase in the number of free-range farms has been noticed
- Microbial diseases cause significant economic losses to pigs by loss in production, by immune suppression, and by increased morbidity and mortality in livestock.





The microbial aggression outcome highly depend on pigs immunity, and medicinal plants, available on the pastures or in the fodder pigs get, could act as immunity enhancers, strengthening the pigs' resistance to diseases.

Aim

Main : To elaborate strategies to improve health and welfare, by the experiments on sows and piglets, conducted on low-input outdoor farms from North Western and Central Romania.

Implicit: Investigating the *in vitro* effects of alcoholic extracts of *Calendula officinalis*, *Thymus vulgaris*, *Cucurbita pepo*, *Coriandrum sativum*, *Allium sativum* on cells responsible for the adaptive immune response in swine raised on a low-input farm from North Western Romania



a-*Calendula officinalis*, b-*Thymus vulgaris*, c-*Coriandrum sativum*, d-*Allium sativum*, e-*Cucurbita pepo*



Chemical composition

<i>Calendula officinalis</i>	<i>Cucurbita pepo</i>	<i>Thymus vulgaris</i>	<i>Coriandrum sativum</i>	<i>Allium sativum</i>
Terpenoids	Carotenoids	Monoterpenes	α-pinene	Aliin
Flavonoides	Phenolic acids	Terpenoids	α-myrcene	Allicin
Quinones	Tocopherols	Flavonoid aglycones	Limonene	Diallyl disulphide
Coumarine	Flavonols	Flavonoids	Citronellyl acetate	Diallyl trisulphide
Volatile oil	Minerals	glycosides,	Geranyl acetate	Ajoene
	Vitamins	Synthetic resin acids	Linalool	Methyl cysteine sulphoxide

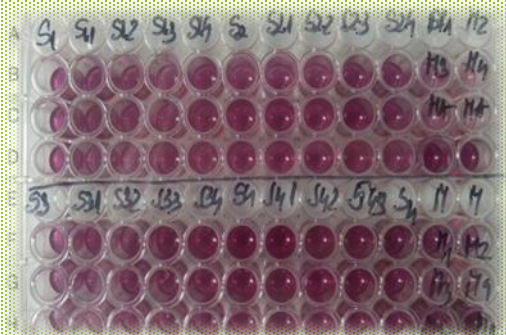


Materials and Methods

The research was carried out on extensively raised Mangalitza **suckling**, **weaned piglets** and **sows** (n=10 for each group).

Blood was sampled during the official campaign for brucellosis testing, with regard to ethical and animal welfare provisions, and subjected to blast transformation test.

- blood was mixed 1:4 with RPMI1640 (Sigma Aldrich, USA), divided in 200µl aliquots in 96 well-plates and
- supplemented with alcoholic plant extracts (*Calendula officinalis*, *Thymus vulgaris*, *Allium sativum*, *Coriandrum sativum*, *Cucurbita maxima*), 1.5 µl/well.



- incubation at 37°C for 48 h, residual glucose was quantified spectrophotometrically (SUMAL PE2, Karl Zeiss, Jena) and glucose consumption was calculated (%).

$$(RPMI\ gluc - sample\ gluc) \times 100 / RPMI\ gluc = SI\%$$

The groups were compared by Student's t test for statistical significance of the results (p<0.05)



Results

The extracts' immunological effects were visible, depending on plant taxonomy, for all variants, when compared with controls (untreated or alcohol treated).

Positive effects were seen mainly for

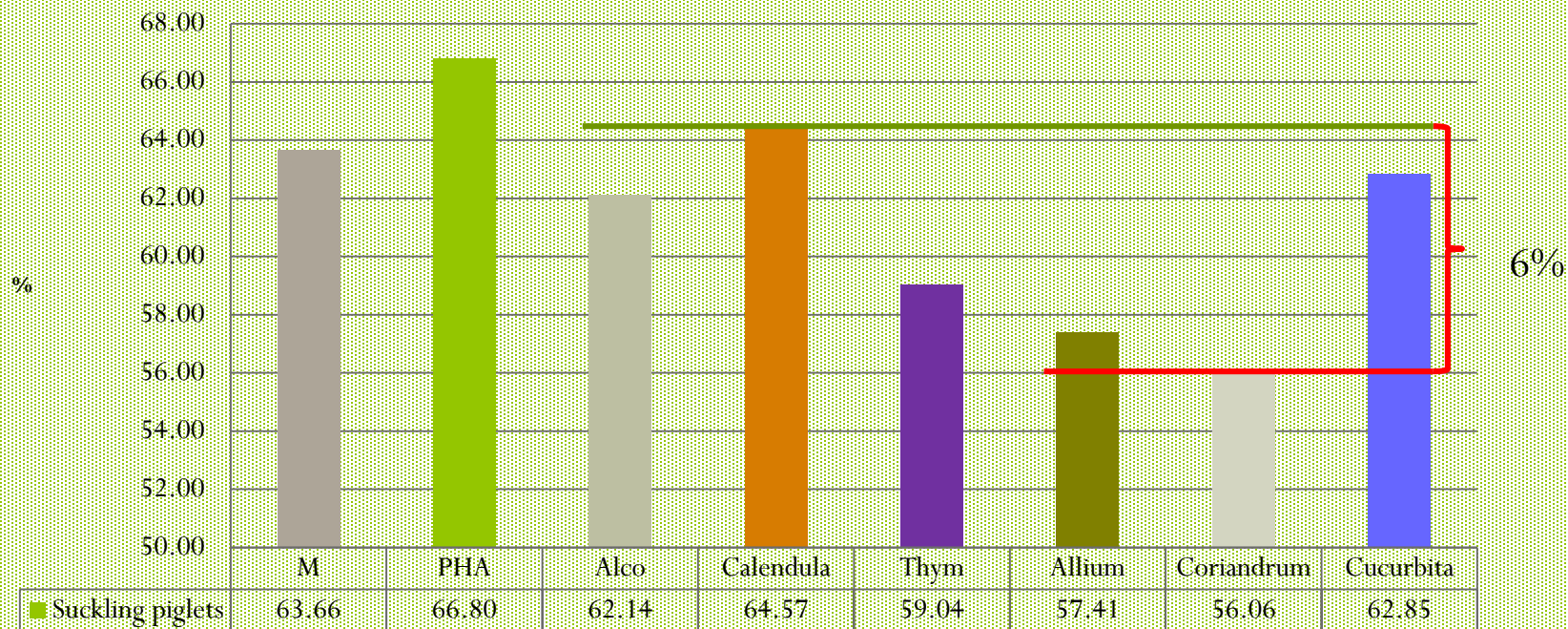
- *Calendula*,
- *Cucurbita* and
- *Allium*, but the extracts' activity was also age dependent.

Calendula (64.47%) and *Cucurbita* (62.85%) worked best in suckling piglets, *Cucurbita* (77.34%) in weaned piglets, none of them stimulating the sow blood cells.



Results

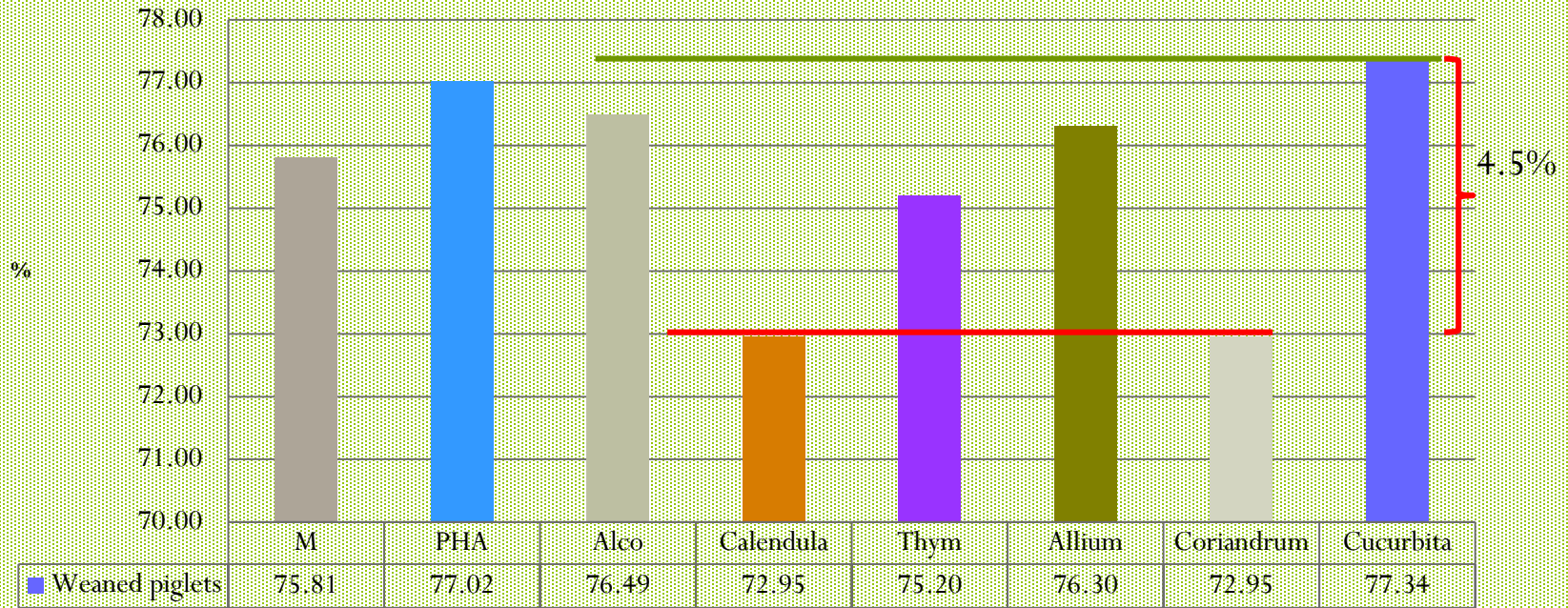
Suckling piglets





Results

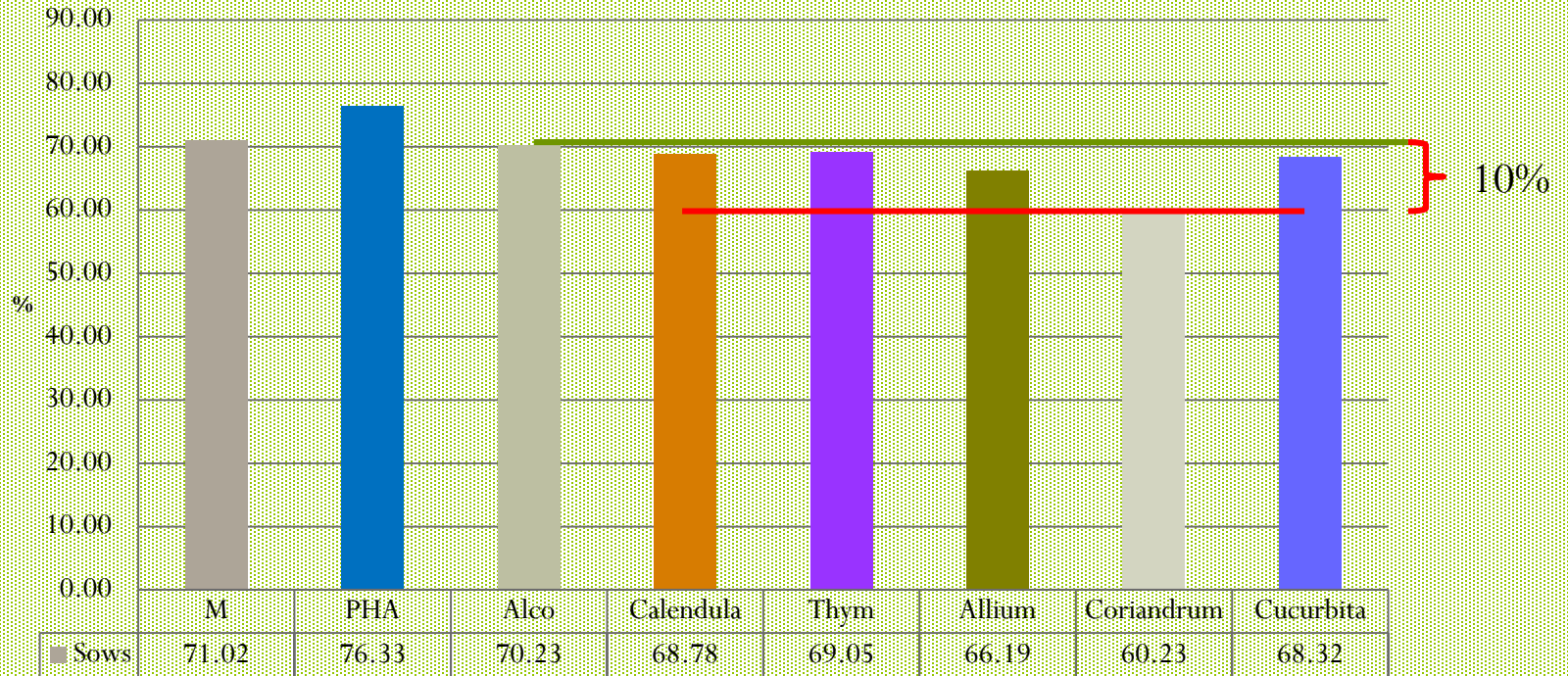
Weaned piglets





Results

Sows





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T test (p values) piglets – weaned piglets

M	PHA	Alco	<i>Calendula</i>	<i>Thym</i>	<i>Allium</i>	<i>Coriandrum</i>	<i>Cucurbita</i>
1.6E-05	1.7E-03	1.6E-06	1.8E-02	2.3E-06	1.6E-06	2.0E-04	2.2E-04

T test (p values) weaned piglets - sows

M	PHA	Alco	<i>Calendula</i>	<i>Thym</i>	<i>Allium</i>	<i>Coriandrum</i>	<i>Cucurbita</i>
0.1982	0.8285	0.0066	0.0359	0.0071	0.0013	0.0003	0.0001

The results indicated statistically significant differences between the young age groups, suckling and weaned piglets ($p=0.017$ to 0.000016) for all plants except marigold, for weaned piglets and sows ($p=0.0001$ - 0.0359) for all plant extract, while for suckling piglets – sows $p=0.0035$ and $p=0.0461$ were recorded for thyme and garlic, respectively.



Conclusion / Discussion: The plant extracts used known for biological effects impacted based on age of the pigs and plant family, proving their immune stimulating capacity.

The immune stimulating activity of the plant alcoholic extracts depended on their taxonomy but also on the age of the pigs (*Calendula* and *Cucurbita* in suckling piglets, *Cucurbita* in weaned piglets, none in sows)



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Thank you!

