Screening of five alcoholic plants extracts effects on the immune status of Romanian EIAV infected horses

Received for publication, March 29, 2011
Accepted, June 9, 2011

POMPEI BOLFĂ*1, CORNEL CĂTOI1, ADRIAN GAL1, MARIAN TAULESCU1, NICODIM FIȚI1, GEORGE NADAS2, MIHAELA NICULAE3, MIRCEA TĂMAȘ4, COSMINA CUC1, MARINA SPÎNU3

1Pathology Department, Faculty of Veterinary Medicine, University of Agricultural Sciences and Veterinary Medicine, Cluj-Napoca, Romania
2Microbiology Department, Faculty of Veterinary Medicine, University of Agricultural Sciences and Veterinary Medicine, Cluj-Napoca, Romania
3Infectious Disease Department, Faculty of Veterinary Medicine, University of Agricultural Sciences and Veterinary Medicine, Cluj-Napoca, Romania
4University of Medicine and Farmacy “Iuliu Hațieganu”, Cluj-Napoca, Faculty of Farmacy

*Corresponding author: Bolfă Pompei Florin1; Phone: +40 264 59 63 84, Fax: +40 264 59 37 92; E-mail: pompeibolfa@gmail.com,
Present adress: Calea Manastur, 3-5, 400372, Cluj-Napoca, Romania

Abstract

Classical medicinal treatments tend to be replaced by alternative therapies, in which the natural plant extracts are being used. In the current study we studied the effects of five alcoholic plant extracts (Calendula flos, Echinacea pallida, Echinacea purpurea, Urtica dioica and Aloe vera) on the dynamics of some immune effectors of equine infectious anemia virus (EIAV) infected horses in vitro. The research focused on the effects of plants active principles on the cellular non-specific defence subsystem and on the functional capacity of the T lymphocyte subsystem. Our findings suggest that the stimulatory effect of Urtica dioica alcoholic extract on the phagocytic function in young, recently EIAV infected horses is followed by an inhibitory effect, that could be explained by the possible rapid carbon particle ingestion, and then by increase cell membrane fragility. Although very closed related taxonomically, the two Echinacea extracts studied here don’t share the same effect in vitro, meaning that different principles are responsible for their in vitro effects. Young horses recently infected with EIAV have an overall increased cellular reactivity compared to older animals, due to infection. On the other hand, due to general increased immunoreactivity in younger animals, the immune recovery effects of the used alcoholic extracts is marked in younger equines.

Keywords: herbal extracts, immunity, immune competent cells, equines, EIAV

Introduction

Nowadays prophylaxis primes over therapy in the majority of diseases affecting man or animals. Numerous studies have been conducted so far that offer scientific proofs on the therapeutic benefits of different plant extracts in the treatment or prophylaxis of different diseases, including some rare forms of cancer (Tajima & Aida, 2000 [1]; Vickers, 2002 [2]). There are herbal plants that have the capacity to modulate different immunologic functions in the pathology of different diseases, but they are also being used as adjuvants in the preparation of vaccines (Florins et al., 2007 [3]; Kaileh et al., 2007 [4]). The capacity of plant extracts or plant isolated active principles to modulate the immune response has been proven by three different pathways: a non-specific stimulation of the immune system
by activating serum complement and PMN neutrophils respectively macrophages, they stimulate phagocytosis (eg. *Echinacea spp.*, *Calendula officinalis* etc.); a second possibility refers to specific stimulation of the immune system, both towards cellular immune reactions and humoral ones, the result of these two directions being antibody synthesis (eg. *Astragalus membraceus*, *Ganoderma lucidum* etc.); the third possibility by which the immune system can be modulated is by a direct influence on immune response by hormonal modulation (via adrenal glands) (BOLFĂ, 2010 [5]).

*Aloe vera* plant (*Aloe barbadensis miller*) has certain components with a low molecular weight are capable of inhibiting the free species of radical oxygen secretion by the human activated neutrophils (HART et al., 1990 [6]). Aloe extract is also used in wound treatment, skin diseases and periodontal disease (WICHTL & BISSET, 1994 [7]).

*Calendula* has a vast applicability in medicine, with application in wound therapy, gastric and duodenal ulcer, skin ulcerations (healing effect) and inflammatory states. Vegetal extracts of *Calendulae flos* present immunomodulatory properties. Phytopharmacological testing with different extracts of *Calendula officinalis* have also indicated that the plant has antiviral properties, anti-HIV and anti/geno-toxic. In vitro, immune-modulating effects were noticed towards human lymphocytes (JIMENEZ-MEDINA et al., 2006 [8]).

By far one of the most widely used immune stimulators of herbal origin, is the *Echinacea* extract. The anti-infectious protection of this plant is most probably ensured by the activating or enhancing properties of the host immune system. Data concerning immune modulating effects induced by *in vitro* administration of this vegetal extract show that these mainly interest the structural elements of non-specific immunity (BUKOVSKY et al., 1993 [9]; BURGER et al., 1997 [10]; JURKŠTIENĖ et al., 2004 [11]; MISHIMA et al., 2004 [12]; RININGER et al., 2000 [13]). Products based on *Echinacea* extracts are indicated for increasing the body nonspecific defence (OLTEANU et al., 2002 [14]). *E. pallida* differs from *E. angustifolia DC*, with which it was initially confused in Europe, through the number of chromosomes, being a tetraploid species (2n=44), unlike *E. angustifolia* which is diploid (2n=22) (BOJAN et al., 2000 [15]).

Biological actions of *Urtica* extracts include diuretic and saluretic effect (elimination of clorurs and ureea). There are numerous studies that prove that the over 100 chemical components of nettles (*Urtica dioica*) are of help for the immune system, but also for the joint system of animals (RANDALL et al, 1999 [16]).

Equine infectious anemia is a persistent viral infection of horses characterized by recurring febrile episodes associating viremia, fever, thrombocytopenia, and wasting symptoms (LEROUX et al., 2004 [17]). The causal agent, EIAV, is a lentivirus (VAN REGENMORTEL et al., 2000 [18]), of the Retrovirus family, with an almost worldwide distribution. EIAV and its associated disease have presented a considerable challenge to veterinary medicine worldwide ever and it is being utilized as an animal model of HIV-1/AIDS research (CRAIGO et al., 2010 [19]). Host immune response to viral infections is complex, involving multiple cellular reactions between distinct cell types (LEROUX et al., 2004 [17]). Cellular immune responses towards EIAV were less characterized then the humoral response (BOLFĂ et al., 2007 [20]). It is well known that cell reservoirs for ungulates lentiviruses include mainly cells from the monocyte-macrophage system. Peripheral monocytosis is observed in horses infected with EIAV (BANKS, 1975 [21]). Tissue macrophages are believed to be the main location for viral replication *in vivo* (BANKS, 1975 [21]; SELLON et al., 1992 [22]). The modality, by which non-specific cellular subsystem is influenced by the infection, can be monitored using the essential function of mature neutrophil, respectively phagocytosis (BOLFĂ et al., 2008 [23]).
In this research we aimed to screen the effects of several alcoholic plants extracts on the dynamics of some immune effectors of EIAV infected horses in vitro. Thus we aimed to monitor the plant effects on the cellular immunity of equids with different ages and at different time periods following the initial diagnosis, compared to the immune system of healthy horses. We focused on two main directions: the effect of EIAV infection on the cellular non-specific defence subsystem and on the functional capacity of the T lymphocyte subsystem was monitored. For our purpose we used five alcoholic plant extracts: Calendula flos, Echinacea pallida, Echinacea purpurea, Úrtica dioica and Aloe vera.

Materials and methods

Biological material.
We used blood collected from 96 horses collected over a period of 2 years from different areas in Transylvania. The horses (54 females and 42 males), were aged between 1 and 25 years, and were divided into six experimental groups, according to disease status, age and period after diagnose. Group 1, formed of 11 EIAV positive horses, aged less than 5 years, less than a year from disease confirmation. Group 2 was formed of 3 EIAV positive horses, aged less than 5 years and diagnosed more than 1 year before. Group 3 was formed of 36 EIAV positive horses, aged more than 5 years, less than a year from disease confirmation. Group 4 was formed of 21 EIAV positive horses, aged more than 5 years, diagnosed more than 1 year before. Group 5 (9 animals) and 6 (16 animals) were control groups, composed of healthy animals, aged less (group 5) respectively more (group 6) than 5 years of age. All equines were clinically healthy, with no clinical sign of disease at the moment of sampling. Horses were used for agricultural works and blood samples were collected always mornings, before the workday began, from the jugular vein. Any haemolysed samples were discarded. Blood samples were analyzed the same day. All horses were previously tested for EIAV seropositivity or seronegativity with the reference agar gel immunodiffusion test (AGID) at the Sanitary Veterinarian Laboratory and for Animal Health Bistriţa, respectively Cluj-Napoca (EIA Kit No. 119, Pasteur Institute, Bucharest, Romania).

Carbon particle inclusion test.
This immunological test was used to monitor spontaneous innate phagocytosis; this activity is in reverse proportion with the optical density of the supernatants. Phagocytic cells engulf inert particles such as carbon due to the defensive capacity of these cells. For this test, we used 3 experimental variants: an alcoholic nettle extract for this experiment, a negative control and an alcohol control variant.

2 microliters (μl) of India ink supernatant, obtained by centrifugation at 6000 rpm for 40 minutes were added to 500 μl heparinised blood and were gently mixed. 150 μl of this mixture was immediately transferred to 2 ml of distilled water, whereas the rest was incubated for 45 min at 37°C, then a second sample was removed, incubated for another 15 minutes and a third sample was removed. The final tubes, with the mixtures of blood, India ink and saline, were centrifuged at 800 rpm, and the supernatants were read spectrophotometrically (λ=535 nm, d=1 cm). There was a decrease in absorbance with time as carbon was phagocytised. The phagocytic activity index was expressed in optical density units (ODU); based on that, the phagocytic activity graph can be built (GHERGARIU et al. 2000 [24]).

Leukocyte blast transformation test.
Mononuclear cells, sensitized in vivo by various antigens, possess the capacity to respond vigorously to the same antigen when contacted in vitro (blast transformation test). In the present study, 160 μl of each heparinised (final concentration of 50 IU/ml) blood sample was mixed with 640 μl of a cell culture medium (RPMI 1640) added with 5% foetal calf serum (FCS), antibiotics (1000 IU penicillin and 1000 μg streptomycin/ml), distributed in
equal aliquots in two wells of a sterile, 96-well plate, for the 8 experimental variants as seen in table 1. For the control (witness) variant we used distilled water. All five plant extracts tested in our experiment represented alcoholic extracts, and were prepared using ethanol.

**Table 1.** Experimental variants used for the blast transformation test (BTT)

<table>
<thead>
<tr>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
</tr>
</thead>
<tbody>
<tr>
<td>Witness 1 μl</td>
<td>Phytomembraglutaminin (PHA) – 1 μl</td>
<td>Alcohol 1,5 μl</td>
<td>Calendula flos 1,5 μl</td>
<td>Echinacea pallida 1,5 μl</td>
<td>Echinacea purpurea 1,5 μl</td>
<td>Urtica dioica 1,5 μl</td>
<td>Aloe vera 1,5 μl</td>
</tr>
</tbody>
</table>

The plates were incubated at 37.5°C in a 5% CO2 atmosphere for 60 hours. Cellular growths can be estimated by calculation of glucose consumption in the medium; this is the reason why at the end of the incubation period, dosing residual glucose allows the calculation of the stimulation indexes. Glucose concentrations were measured in the initial medium and in all the supernatants using a standard (100 mg/dl) glucose solution and a colorimetric test. After mixing 12.5 μl of the culture supernatant with 0.5 ml ortho-toluidine, the samples were boiled for 8 min, quickly cooled in cold tap water, and read spectrophotometrically at a wavelength of 610 nm (SUMAL PE 2) in 96-well plates using the reagent as a blank. The transformation index (TI %) was calculated as follows: TI= [(MG-SG)/ MG] x100, where MG is the glucose concentration in the initial culture medium, and SG the glucose concentration in the sample after incubation (GHERGARIU et al. 2000 [24]).

**Plant extracts.**

1. Fluid *Calendulaceae flos* extract: We used *Calendula officinalis* inflorescence, from the experimental field of UASVM Cluj-Napoca. Dried inflorescences were grinded to a fine powder (VI FR X sieve), followed by the preparation of a fluid extract 1:2 using 60˚ alcohol, using Squibb’s repercolation technique (IONESCU STOIAN, 1977 [25]). Thus, from 1 part of vegetal product, we obtained 2 parts of extract (m/m). The extract was standardized in flavonoids content, expressed in rutoid (0.15%) and had a residue through evaporation of 12.3%. The content in total flavonoids, was determined using a spectrophotometric method (ROMANIAN FARMACOPEIA, 1993 [26]). The vegetal product *Calendulaceae flos* contains flavonoids, mucilage, saponins, carotenoids, free triterpene, phenolcarboxilic acids and immunostimulatory polysaccharides. EUROPEAN PHARMACOPEA (2008) [27] stipulates a content of minimum 0.4% flavonoids, evaluated spectrophotometrically. TÂMAŞ et al. (1997) [28], determined a content of 0.3 – 0.35% flavonoids in the Romanian product *Calendulaceae flos*.

2. Fluid extract (1:1) of *Echinacea palida* (Nutt.), Nutt. (Asteraceae): For the preparation of this extract, we used integral vegetal product (roots, stems, leaves and inflorescence), harvested during blooming period (July), from the Experimental field of USAVM Cluj-Napoca, in the III-rd year of cultivation. The vegetal product was washed from soil, fragmented and dried, then grinded to a fine powder (VI FR X sieve). The fluid extract (1:1) was obtained using repercolation technique with 60˚ alcohol. This extract presents a residuum by evaporation of 8.71%, content in total polyphenols of 3.73% and was standardized to a content of 1% immunostimulant polysaccharides, using the spectrophotometric technique indicated by TÂMAŞ M. et al. (1991) [29]. The content in polyphenols was determined using the spectrophotometric technique with Folin-Ciocalteu reactive, according to PH. EUR. (2008) [27], and expressed in galic acid equivalents. *E. pallida* contains immunostimulatory polysaccharides (heteroglicans), phenol-carboxilic compounds, from which echinacozid is the most important quantitatively, poliine, poliacetilene, volatile oil and flavonoids. PH. EUR. (2008) [27] stipulates a content of 0.2%...
echinacosid in the roots of *E. pallida*, determined by High Performance Liquid Chromatography (HPLC).

3. Fluid extract of *Echinacea purpurea* (L.) Moench: For the obtaining of *Echinacea purpurea* fluid extract, we used the product *Echinacea purpurea herba*, which is the aerial part of the plant, harvested in the III-rd year of cultivation from the Experimental field of USAVM Cluj-Napoca. The vegetal material was obtained using Squibb’s repercolation technique, with 60° alcohol. *Echinacea purpurea* extract has a residuum by evaporation of 5.52% and was standardized in total polyphenols (2.35%) expressed in galic acid equivalents. Quantitative determination of total polyphenols was made using the spectrophotometric technique with Folin-Ciocalteu reactive, (PH. EUR., 2008 [27]). PH. EUR. (2008) [27] stipulates a content of 0.1% phenolcarboxylic acids for *E. purpurea*, determined by HPLC.

4. *Urtica dioica* extract: We used for our experiment an alcoholic (60°) extract, from the aerial part of *Urtica herba* with stems and leaves. This plant contains flavonoids, chlorophyll, free amino acids, vitamins (C, B and K), triterpene and sterols, mineral salts (K, Si and nitrates), organic acids (citric, formic and acetic). There is still a debate on the presence of glucochinins responsible of the antidiabetic effect. *Urtica* brushes contain amines (histamine, coline, serotonine), responsible for the neetle’s rash (WICHTL & BISSET, 1994 [7]).

5. *Aloe barbadensis* miller (*Aloe vera*). alcoholic extract: We used an alcoholic (60°) extract obtain from *Aloe* powder. This has a brown colour and is soluble in ethanol (at warm), partially soluble in water, and not soluble in ether and chloroform. *Aloe* (the product), contains antracenozids (15-40%), in which aloin is the main component and also rezin (0.01-0.02%).

Statistical analysis.

Our results were expressed as mean ± standard deviation. The results were analyzed for normality of distribution using the Shapiro–Wilk normality test. The majority of data sets were found not to be normally distributed. Mean comparison was done by the Kruskal-Wallis test followed by the Wilcoxon test; a confidence level of 95% (p < 0.05) was considered significant. To assess the influence of different plant extracts on the immune cells we compared the results from seropositive horses (infected <1 year or >1 year before the experiment) with those from healthy animals of the same age. Different age groups of EIAV positive and negative horses were as well compared, to determine the influence of extracts related to the age of the animals. The R free software package was used for all statistical analyses.

Results and discussions

The results of the phagocytosis test are illustrated below (figure 1, 2, 3 and 4). Phagocytic activity can be modified in a positive (stimulatory) or negative (inhibitive) way by certain vegetal extracts.

![Figure 1. Boxplot diagram for the phagocytic activity 1](image)

![Figure 2. Boxplot diagram for the phagocytic activity 2](image)
activity 1 (0-45 min.) of the experimental variants treated with alcohol and nettle alcoholic extract

activity 2 (45-60 min.) of the experimental variants treated with alcohol and nettle alcoholic extract

Figure 3. Boxplot diagram for the phagocytic activity 1 (0-45 min.) of the witness variant and the nettle alcoholic extract treated variant

Figure 4. Boxplot diagram for the phagocytic activity 2 (45-60 min.) of the witness experimental variant and the variant treated with nettle alcoholic extract

Following the analysis of our results, the phagocytic activity was enhanced in the second monitored period (from 45 to 60 minutes), proving a delayed in vitro activation of phagocytic blood cells from the investigated horses. Regarding young, recently infected horses (group 1), the reactive capacity of phagocytes is significantly raised in the first period (0 to 45 minutes), followed by an inhibition in the second period (p<0.05). The alcoholic extract of *Urtica dioica* proves itself to be more of an inhibitor of phagocytosis in the first period, the effect being more intense in the age category of under 5 years old (group 5) of healthy horses (p<0.05), followed by a stimulatory effect in the second period.

In young horses, EIAV positive, aged less than 5 years, having under one year from the disease diagnostic (group 1), the alcoholic *Urtica dioica* extract demonstrates a stimulatory effect on phagocytosis in the first stage, followed by an inhibitory effect (p<0.05).

*Urtica dioica* extract seems to have an inhibitory effect on first stage phagocytosis in infected horses aged over 5 years old, regardless of the interval passed from the diagnosis (group 3 and 4) and in the ones with ages below 5 years where more than 1 year has passed since the diagnosis (group 2); this initial effect is followed in the second period by a slight stimulatory effect. In EIAV infected horses aged over 5 years old, and more than 1 year from initial diagnosis (group 4), in the case of the untreated and alcohol treated groups, phagocytic activity is significantly enhanced in the second period (the 45-60 minutes interval) compared to the first interval (0-15 minutes), which suggests that in this category of animals, the reactive capacity of the phagocytes is still preserved (p<0.05).

*In vitro*, the *Urtica dioica* alcoholic extract treatment aiming to quantify the phagocytic activity follows a similar model to spontaneous phagocytosis. By looking at the *Urtica dioica* extract effects on cells isolated from healthy horses, during the first reading period, one might state that it acts rather as an inhibitor. This change during the second period, when the effect of the alcoholic extract becomes predominantly stimulatory.

If we compare our results with those obtained by BOLDIZSAR (2001) [30] (mainly the first monitored interval- respectively the first 45 minutes of incubation), we observe that they share some common aspects, but there are also certain differences regarding phagocytic activity. Thus, similar to Boldizsar’s results (BOLDIZSAR, 2001 [30]), in EIAV positive horses, phagocytic activity drops with ageing of animals and with the amount of time that
passes from initial diagnosis. On the other hand, the more time passes from sample incubation, phagocytic activity seems to get back to normal (during the 45-60 minute interval).

Comparative evaluation of the **lymphoblastic transformation test** in horses (figure 5, 6, 7, 8, 9, 10, 11 and 12), reveals several differences between animal groups. Thus, in the case of the healthy animals, independently of age, the stimulation indices values maintain themselves in all experimental variants to approximately the same level (there are no statistic differences between the eight experimental variants that were applied). Similarly, in seronegative animals, independent of age, we observed much lower values of the stimulation indices compared to seropositive animals, regardless of age or diagnostic date, probably because of the absence of antigenic stimulation *in vitro*. Therefore, in the equine control groups (group 5 and 6) (excepting the untreated witness group) and in the EIAV positive horses, more than one year following diagnostic (group 2 and 4), the response capacity of lymphocytes to several stimuli of the experiment drops with ageing, confirming the balance that exists between virus and the animal’s immune system in the asymptomatic evolution stage of the disease. In seropositive horses, less than one year from diagnosis (group 1 and 3), the situation is reversed: the response capacity of lymphocytes to several stimuli in the experiment is enhanced in the older animals (group 3) (with the exception of *Calendula, Echinacea pallida and Echinacea purpurea*).

The response capacity to the mitogen inductors used is marked by the evolution of the disease in animals aged over 5 years old and seropositive (group 4). This could be explained by the lack of circulating virus in the infected animals’ blood, as opposed to the tendency seen in young animals (up to 5 years old – group 3), where the response to certain mitogens is enhanced by the more time passed from the initial diagnostic.

There are no statistically significant differences in the stimulation indices between the 8 working variants (the cell immune reactivity is not altered, just as in healthy horses) in horses aged up to 5 years old and that have less than one year since EIAV infection diagnosis (group 1). Unlike that, in horses aged up to 5 years, with more than one year from diagnosis (group 2), among the utilized immune modulators, *Aloe vera* (*p*<0.05) PHA M, and *Calendula* produced a significantly increased stimulation compared to *Echinacea purpurea* (with a rather inhibitory effect).

In group 4 the effect of *Echinacea purpurea* is inhibitory, being lower compared to the witness, alcohol and PHA M variants.

![Figure 5](image1.png)  
**Figure 5.** Boxplot diagram for the stimulation indexes of the witness experimental variant

![Figure 6](image2.png)  
**Figure 6.** Boxplot diagram for the stimulation indexes of the variant treated with Phytohemaglutinin M
Figure 7. Boxplot diagram for the stimulation indexes of the alcohol – witness experimental variant

Figure 8. Boxplot diagram for the stimulation indexes of the variant treated with *Calendula* extract

Figure 9. Boxplot diagram for the stimulation indexes of the variant treated with *Echinacea pallida*

Figure 10. Boxplot diagram for the stimulation indexes of the variant treated with *E. purpurea*

Figure 11. Boxplot diagram for the stimulation indexes of the variant treated with *Urtica dioica*

Figure 12. Boxplot diagram for the stimulation indexes of the variant treated with *Aloe vera*

An inhibitory effect of *Echinacea purpurea* extract is visible in group 3 (*p*<0.05) compared to the witness group, the alcohol-group, PHA M, nettle and aloe, but also compared to *Calendula* variant. The immune recovery effects of the used extracts are seen mainly in young animals; these effects are occur in horses aged up to 5 years, with less than one year from the EIAV diagnosis (group 1), after cell treatment with *Calendula, Echinacea pallida* and *Echinacea purpurea*; immune recovery occurred in the same age category but having more than one year from diagnosis (group 2) following the treatment with *Aloe vera, Urtica*
 dioica and Calendula extract; in group 4, only the Echinacea pallida extract has registered higher transformation indices than the two control variants ($p>0.05$).

Even though we tested two very similar plants taxonomically (Echinacea pallida and Echinacea purpurea), which belong to the same genre, the in vitro effects of their alcoholic extracts are different. This demonstrates that the two plants have different active principles, for which out tested cells do have more or less receptors. Chemical composition and biological properties of Echinacea purpurea, are similar with those of E. pallida, but the first one has an increased content of choric acid and contains no echinacozide, which is specific for E. pallida. The polysaccharides of the two species are different structurally, as well as their composition in volatile oils.

In a review focused on different experimental studies, MELCHART et al. (1994) [31] have proved the modulating effects of the immune parameters of Echinacea angustifolia, suggesting that further study should continue and investigate it. Focusing on the same directions, BUKOVSKI et al. (1993) [9], BURGER et al. (1997) [10], MISHIMA et al. (2004) [12], RININGER et al. (2000) [13], have suggested that plant extracts of Echinacea species are capable of inducing an increased blast stimulation level. This tendency was confirm by our study. In addition, it is proven that Echinacea purpurea does not have the same active principles and does not have the same beneficial effects in vitro.

Most recent studies indicate that multiple Prunella vulgaris constituents have profound anti-viral activity against EIAV, providing additional evidence of the broad anti-viral abilities of plant extracts. The ability of the aqueous extracts to prevent entry of viral particles into permissive cells suggests that these extracts may function as promising microbicides against lentiviruses (BRINDLEY et al., 2009 [32]).

Conclusions

Due to a lack of antigenic stimulation in vivo, the value of stimulation indexes are decreases in healthy compared to EIAV infected equines, regardless of age or duration of disease progression. The stimulatory effect of Urtica dioica alcoholic extract on the phagocytic function in young, recently EIAV infected horses is followed by an inhibitory effect which could be explained by the possible rapid carbon particle ingestion, followed by increase cell membrane fragility. Although very closed taxonomically, Echinacea pallida and Echinacea purpurea, their alcoholic extracts do not share the same effects in vitro, proving that different principles are responsible for the well known beneficial effects of the two plants. Young animals which are recently infected show increased cellular reactivity compared to older animals, due to infection. On the other hand, general increased immunoreactivity in younger animals makes the immune recovery effects of the used alcoholic extracts to be more obvious in this category.

References