Animal Feed Science and Technology 257 (2019) 114265

Contents lists available at ScienceDirect

Animal Feed Science and Technology

journal homepage: www.elsevier.com/locate/anifeedsci

# Addition of oregano or green tea extracts into the diet for Jersey cows in transition period. Feeding and social behavior, intake and health status. Plant extracts for cows in the transition period

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ARTICLE INFO

*Keywords:*
Diseases
Disorders
Intake
Plant extracts
Transition period

ABSTRACT

This study aimed to evaluate the effects of feeding oregano or green tea extracts on feeding and social behavior, intake and health status of dairy cows during the transition period. Twenty-four Jersey cows were randomly assigned to treatments: control (CON), 10 g / day of oregano extract (OE) or 5 g / day of green tea extract (GT). Dry matter intake (DMI) was evaluated from 7 days pre-calving until the 16 days of lactation using chromium oxide. Feeding and social behaviors as well as the occurrence of health problems were monitored throughout the experimental period. Plant additives did not affect dry matter intake before calving, but after calving Oregano supplementation tended to increase DMI in 1.3 kg compared with Control. Before calving, during the daytime period, OR tended to decrease time spent lying down in 38 min and eating the concentrate in 8.6 min compared with GT. After calving, Oregano reduced time spent eating the concentrate in 9.8 and 7.5 min compared with GT and CON, respectively. The number of total visits to the feeder was lower for cows supplemented with OE compared to the others. Before calving, when GT was added to the diet, cows visited 3.3 times more the feeder and tended to interact 1.2 and 1.6 times less compared to cows in OE and CON groups, respectively. The number of metabolic disorders and clinical and subclinical infectious diseases was similar between treatments, but cows in OE group that were affected by some disease or disorder, tended to present clinical symptoms or subclinical diagnosis on average 8 days later than those supplemented with GT. Oregano and green tea extracts administered into the diet of lactating Jersey cows during the transition periods change several traits of feeding and social behavior but do not change diet intake and the occurrence of diseases.

## 1. Introduction

The use of plant extracts as feed additives for dairy cattle have been increasing due to the concerns about human health and

*Abbreviations:* CON, control; OE, oregano extract; GT, green tea extract; FP, faecal production; TDMI, total dry matter intake; FDMI, forage dry matter intake; CIR, concentrate intake rate; SIR, silage intake rate

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https://doi.org/10.1016/j.anifeedsci.2019.114265
Received 3 April 2019; Received in revised form 19 August 2019; Accepted 23 August 2019

animal welfare (Maciej et al., 2016). Catechins and carvacrol are the main secondary metabolites produced by green tea and oregano plants, respectively (Manach et al., 2004; Oh et al., 2017). In humans and monogastric animals, both compounds have been used primarily as antibacterial, antidepressant as well as powerful immune system and antioxidant stimulants (Aristatile et al., 2015; Cyboran et al., 2015; Gonçalves et al., 2015) while in ruminants, they are mostly used to modulate ruminal fermentation (Cobellis et al., 2016; Oh and Hristov, 2016; Kolling et al., 2018).

The digestive process of ruminants alters the amount and biochemical structure of the secondary compounds of plants that reach the bloodstream (Gladine et al., 2007). Previous studies indicate that they do not lose their functionality and, in ruminants, may stimulate the immune defense and the antioxidant systems (Winkler et al., 2015; Maciej et al., 2016). However, this ability has been little explored during the transition period, when physiological and metabolic changes have great impact in the defense system of the organism, making dairy cows more susceptible to the occurrence of metabolic disorders and infectious diseases (Trevisi et al., 2012; Garro et al., 2014).

Some plant secondary compounds may also alter the expression of the serotonin and dopamine's receptors (Trabace et al., 2011; Zotti et al., 2013; Mirza et al., 2013), modifying the response of the animal to the environment. Some alterations in feeding behavior like the time spent ingesting the concentrate and engaged in social behavior e.g. those related to agonist interactions have already been observed in dairy heifers (Gabbi et al., 2009a; Kolling et al., 2016), but still have not been reported for dairy cows, especially during the transition period.

The effects of plant extracts on the immune system has also been evaluated (Gladine et al., 2007; Gabbi et al., 2009b), but its potential in reducing the occurrence of metabolic disorders and infectious diseases when fed during the transition period is still little explored. Our hypotheses are 1) green tea or oregano extracts modify feeding and social behavior of cows during the transition period; 2) green tea or oregano extracts reduce the occurrence of metabolic disorders and infectious diseases during the transition period. The objective of this research was to evaluate the intake, feeding and social behavior and health status of dairy cows fed with oregano and green tea extracts during the transition period.

## 2. Material and methods

### 2.1. Location and duration

This study was approved by the Ethics Committee for the Use of Farm Animals of the Universidade Federal do Rio Grande do Sul, protocol number 29838. The study took place during the hot season at Embrapa Clima Temperado, Capão do Leão, Rio Grande do Sul, Brazil between October 2015 and January 2016. The climate of the region is classified by Köppen as subtropical humid. The data of air temperature (ºC), relative air humidity (%), wind speed (km / h) and precipitation (mm / day) during the study were collected at the meteorological station of the Agrometeorology Laboratory of Embrapa Clima Temperado, Capão do Leão, Rio Grande do Sul, Brazil. The mean values of air temperature, relative air humidity and wind speed during the study were respectively (mean ± SEM) 20.1 ± 0.5 °C, 85.3 ± 1.6% and 14.2 ± 0.9 km / h, and the cumulative rainfall during the whole experimental period was 847 mm (Table 1).

### 2.2. Animals

Twenty-four Jersey cows, selected from the experimental herd according to the number of parturitions, body weight (BW) and body condition score (BCS), were enrolled at 28 before their expected calving date (day -28) and stayed in the trial until 21 days of lactation (day +21). The first 7 days were destined for the adaptation to the diet, and the remaining 42 days were destined for measurements. Before the trial, for about 32 days, which comprised the first five weeks of the dry period, all cows grazed rangeland pasture improved with annual ryegrass (*Lolium multiflorum*), with free access to water and natural shade provided by trees of several species.

### 2.3. Daily routine and nutritional management

During the 28 days prior to calving, all cows were confined in a free stall barn with rubber mattress, in a proportion of stall to cows of 1:1, and received a diet composed of 4.6 kg of DM per cow per day of corn silage and 3.9 kg of DM per cow per day of

**Table 1**
Mean and amplitude of air temperature (ºC), relative air humidity (%), wind speed (km / h) and precipitation (mm / day) during the experimental period.

| Month | Temperature | | Humidity | | Wind speed | | Precipitation |
|---|---|---|---|---|---|---|---|
| | Mean | Amplitude | Mean | Amplitude | Mean | Amplitude | |
| October | 16.3 | 21.1 – 4.1 | 88.9 | 97.0 – 73.3 | 35.1 | 54.7 – 24.1 | 315.2 |
| November | 18.9 | 22.6 – 15.6 | 84.3 | 96.4 – 72.0 | 8.6 | 17.0 – 3.5 | 192.3 |
| December | 22.1 | 25.3 – 17.3 | 83.7 | 94.9 – 52.8 | 6.9 | 12.8 – 0.0 | 262.0 |
| January | 23.2 | 25.1 – 21.1 | 84.5 | 95.9 – 70.8 | 6.2 | 13.4 – 2.0 | 77.5 |

**Table 2**
Ingredients and chemical composition of the diets fed to the cows during the transition period.

| Item[2] | Pre-calving diets[1] | | | Post-calving diets[1] | | |
|---|---|---|---|---|---|---|
| | CON | GT | OE | CON | GT | OE |
| Corn silage | 54.1 | 54.1 | 54.1 | 39.5 | 39.5 | 39.5 |
| Pasture | – | – | – | 19.8 | 19.8 | 19.8 |
| Ground corn grain | 19.7 | 19.7 | 19.7 | 24.7 | 24.7 | 24.7 |
| Wheat bran | 8.2 | 8.2 | 8.2 | 7.8 | 7.8 | 7.8 |
| Soybean meal | 8.2 | 8.2 | 8.2 | 3.0 | 3.0 | 3.0 |
| Ground soybeans | 7.3 | 7.3 | 7.3 | 1.9 | 1.9 | 1.9 |
| Pre-calving mineral salt | 2.4 | 2.4 | 2.4 | – | – | – |
| Mineral and vitamin supplement | – | – | – | 1.9 | 1.9 | 1.9 |
| Urea | – | – | – | 1.0 | 1.0 | 1.0 |
| Common salt | – | – | – | 0.2 | 0.2 | 0.2 |
| Calcium carbonate | – | – | – | 0.3 | 0.3 | 0.3 |
| OE | – | – | 5.6 | – | – | 5.6 |
| GT | – | 2.8 | – | – | 2.8 | – |
| | Pre-calving diets | | | Post-calving diets | | |
| CP | 14.7 | | | 15.0 | | |
| NDF | 50.8 | | | 35.6 | | |
| DMIVD | 72.0 | | | 73.1 | | |

[1] Diets: CON = control; GT = 5 g / day of green tea extract; OE = 10 g / day of oregano extract.
[2] Item: CP = crude protein; NDF = neutral detergent fiber; DMIVD = dry matter in vitro digestibility.

concentrate. Concentrate was composed of 43.0% of ground corn grain, 17.8% of wheat bran, 17.9% of soybean meal, 16.0% of ground soybeans and 5.3% of pre-calving mineral salt (Table 2) in order to meet nutritional requirements of cows with 440 kg of BW at 260 days of gestation (National Research Council (NRC, 2001). During this period, the diet was given twice a day (0700 h and 1600 h).

From calving to 21 days of lactation, the cows grazed rangeland pasture improved with annual ryegrass (*Lolium multiflorum*) during the day (between morning and evening milking) and were confined in a barn with rubber mattress stalls at night in a proportion of stall to cows of 1:1 (after the evening milking until the morning milking). Cows were fed approximately 4.0 kg of DM per cow per day of corn silage and 4.1 kg of DM per cow per day of concentrate composed of 61.0% of ground corn grain, 19.5% of wheat bran, 7.3% of soybean meal, 4.9% of ground soybeans, 4.9% of mineral and vitamin supplement, 0.5% of common salt, 0.7% of calcium carbonate and 2.4% de urea (Table 2), in order to meet nutritional needs considering cows with 400 kg of BW, at the beginning of lactation and producing 17 kg of milk with 3.6% fat (National Research Council (NRC, 2001). After calving, the concentrate was individually supplied twice daily before each milking (0700 h and 1600 h) and corn silage was individually supplied after milking (1800 h). The cows were milked at 0730 h and 1630 h. Pasture samples were collected weekly by grazing simulation hand plucking (Euclides et al., 1992) and along with samples of pre and post-calving diets, ingredients were analyzed for DM (Detmann et al., 2012, INCT-CA nº G-003/1), CP (Kjedahl method, nitrogen content was then multiplied by 6.25 to calculate CP) (Association of Official Analytical Chemists (AOAC, 2011), NDF (Van Soest et al., 1991) and DMIVD (Association of Official Analytical Chemists (AOAC, 2011; Table 2).

## 2.4. Experimental design

Cows were randomly divided in three similar groups (in terms of number of parturitions, BW and BCS) with repeated measures in time (days) and eight animals each, being: Control (CON) - without addition of plant extracts in the diet, Oregano Extract (OE) - addition of 10 g per cow per day of oregano extract (*Origanum vulgare*) in diet and Green Tea Extract (GT) – addition of 5 g per cow per day of green tea extract (*Camellia sinensis* L.) in diet.

At the start of the study, cows in the CON treatment averaged ( ± SEM) 3.0 ± 2.2 lactations, 444 ± 10 kg of body weight (BW) and 3.3 ± 0.5 points of BCS; cows in the OE treatment averaged ( ± SEM) 2.7 ± 1.8 lactations, 420 ± 42 kg of BW and 3.6 ± 0.2 points of BCS and cows in the GT treatment averaged ( ± SEM) 2.5 ± 1.5 lactations, 459 ± 31 kg of BW and 3.6 ± 0.2 points of BCS.

Plant extracts were given to the animals as powder, homogenized in 0.5 kg (as fed) of concentrate top dressed onto the concentrate at the time of morning feeding. The oregano extract (Orego Stim® - Meriden Animal Health Ldta. - Northampton, UK, redistributed in Brazil by Advet Animal Nutrition containing 5% of essential oil of oregano plants (*Origanum vulgare* subsp. *Hirtum*) and 95% of inert substance), had a minimum concentration of 50 g / kg containing 80–82% of Carvacrol, 2.5–3.0% of Tymol, 3.5–9.0% of p-Cymene and 2.0–5.0% of Y-Terpinene. The green tea extract had a concentration of approximately 56% ( ± 2.5%) of polyphenols.

### 2.5. Performance and physiological characteristics

Milk production (kg) was recorded every day, at the morning and evening milking during the experimental period using DeLaval milk meter MM25. Animals were weighed weekly without previous fasting after milking in the morning but before feed was offered. Every week, after milking each cow was awarded a body condition score on a scale of 1 (very poor) to 5 (grossly fat) (Edmonson et al., 1989).

### 2.6. Dry matter intake

Fecal production was estimated using 10 g / cow per day of chromium oxide ($Cr_2O_3$). The marker was provided starting on day -12 and fecal collections were performed from day -7 until day +16 in relation to calving day. The $Cr_2O_3$ was ministered mixed to 0.5 kg of concentrate during the morning feeding, and fecal samples collected twice a day, before each milking (Kozloski et al., 2006).

The concentration of chromium in the dry faeces was determined by atomic absorption spectrophotometry (method with flame, AANALYST, PerkinElmer, Waltham, EUA), and the samples were previously digested using nitro-perchloric acids (Tedesco and Volkweiss, 1995). Faecal production (FP) was calculated as according to Pond et al. (1989).

Total dry matter intake (TDMI; g / day) was calculated as: TDMI = (DM total fecal)-(DMI concentrate*(1-digestibility of concentrate))/(1-forage digestibility) and the forage dry matter intake (FDMI; g / day) was calculated as: FDMI = (DM total fecal)/(1-forage digestibility). The in vitro digestibility of concentrate and forage was performed according to AOAC (2011).

The feeding efficiency and feeding efficiency for milk corrected energy were calculated dividing the milk production and milk corrected energy production by DMI, respectively. Milk production data was corrected for energy (energy corrected milk - ECM), using the formula ECM = [(0.3246*kg milk) + (12.86*kg fat) + (7.04*kg true protein)], true protein calculated as 95% of the milk crude protein (National Research Council (NRC, 2001) and milk fat and protein content determined by infrared spectrophotometry (Fonseca and Santos, 2000).

### 2.7. Behavior measurement

The daily behaviors rumination, activity and rest were evaluated continuously throughout the experimental period (Table 3). Briefly each cow was fitted with an individual behavior logger on a neck collar (C-tech/Chip Inside Engineering and Technology, Santa Maria, Brazil). Data was stored in an internal memory and further transferred daily by radiofrequency to the management software (Healthy Cow Manager®) when cows passed close to portal installed at the exit of the milking parlor.

Other feeding behaviors such as time spent at the feed trough with and without ingestion, time spent eating the concentrate and silage, concentrate and silage intake rates and the time spent grazing, standing up and lying down (Table 3) were assessed by visual and focal observation of each cow at 10-minute intervals (Martin and Baterson, 1993).

Number of events of water intake, visits to the feed trough with and without ingestion, licking the trough, aggression events (aggressive behavior when disputing resources such as food and water) and non-aggressive interactions between animals as well as duration of each visit to the trough with food ingestion were observed continuously during the pre-calving period and recorded whenever they occurred.

During the entire experiment, the animals of the different treatments had free access to water and to shade throughout the

**Table 3**
Ethogram describing the evaluated behavioral activities.

| Behavioral characteristics | Description of activities |
|---|---|
| Rumination time | Time spent with regurgitation, chewing and reingestion of the digesta boluses (minutes) |
| Rest time | Time spent without presence of mandibular movements (minutes) |
| Activity time | Time spent on movement and displacement (minutes) |
| Standing up time | Time spent standing (minutes) |
| Lying down time | Time spent with the flank in contact with the floor (minutes) |
| Time spent in the feed trough | Time spent sorting, apprehending, chewing and ingesting the food (minutes) |
| Grazing time | Time spent seizing and chewing fresh pasture during grazing activity (minutes) |
| Time spend eating the concentrate or silage | Effective time spent ingesting the concentrate or silage without pauses (minutes) |
| Concentrate or silage intake rate | Amount in grams of MS of concentrate or silage consumed per minute of the time of ingestion |
| Number of visits to the trough | Total number of times that the cows entered their head into the feeding space of the trough |
| Number of visits to the feeder with food ingestion | Number of times that the cows entered their head into the feeding space of the trough and consumed food |
| Number of visits to the feeder without food ingestion | Number of times that the cows entered their head into the feeding space of the trough but without food consumption |
| Duration of each visits to the trough with food ingestion | Mean time spent at each visit to the troughs with food consumption (minutes) |
| Licking the trough | Number of events in which the cows licked the trough |
| Social interactions | Number of positive contact between animals (e.g. approaching without aggressive displacement or butting) |
| Aggressive interactions | Number of actions of intimidation or confrontation with other animals e.g., aggressive displacement resulting in the withdrawing from the feed or stall and butting) |

experiment. The behavioral activities performed during the milking and the displacement between the free-stall and the milking parlor were not evaluated.

### 2.8. Health condition

The occurrence of clinical and subclinical health problems was monitored and registered throughout the experimental period. Placental retention was defined as the inability to expel the placenta within 24 h post-calving (Sandals et al., 1979). Clinical mastitis was characterized by the presence of abnormal milk (clot formation) and visible signs of inflammation such as redness and swelling of the udder. Metritis was assessed on the observation of vaginal discharge (Huzzey et al., 2007) and lameness was accessed with locomotion score during the displacement between the barn to the milking parlor, using the 5 point numerical classification system, where 1 = healthy, 5 = severely lame (Flower and Weary, 2006).

Blood samples were obtained from each cow via jugular venipunction using 5-ml tubes (Vacutainer; Becton-Dickinson, Rutherford, NJ) without anticoagulant on days -7, -4, -2, 0 (calving day), 2, 4, 7, 14, 21, before the morning feed delivery. Further blood samples were centrifuged at 2000 *g* for 10 min and the serum was used to determine calcium concentration using commercial kit (Ca Arsenazo Liquiform, colorimetry, Labtest Diagnóstica). Cases of subclinical and clinical hypocalcaemia were considered when the serum calcium concentration ranged from 5.5 to 8.0 mg / dL and below 5.5 mg / dL, respectively (Goff, 2008). The concentration of circulating β-hydroxybutyrate (BHBA) was determined using a digital autoanalyzer (Free Style/Abbott). The thresholds used for the diagnosis of subclinical and clinical ketosis were blood concentrations of BHBA ≥ 1.2 mM and ≥ 3 mM, respectively (Garro et al., 2014).

The total number of diseases and metabolic disorders was calculated as well as the period of time elapsed from calving until the diagnosis of the first disease or clinical or subclinical disorder.

### 2.9. Experimental design and statistical analysis

Continuous data were tested for normality (PROC UNIVARIATE; SAS®) using *Shapiro-Wilk* test. Differences in DMI, behavior parameters and health condition between CON, GT and OE groups were analyzed using the MIXED procedure (SAS® version 9.4, SAS Institute, Cary, NC). The statistical model included the fixed effects of treatment (n = 3, CON, GT and OE), days of evaluation and your interaction (treatment by days). Animal and the residue were considered as the random effects. A structural selection test was performed using the Bayesian information criterion (BIC). Covariance structures tested were compound symmetry, first-order autoregressive, toeplitz, and unstructured. Parity, BW and BCS measured at the beginning of the study (but before treatments were started) were included in the model as covariates. Analysis of variance was performed to test the effect of the interactions. The contrasts CON x OE, CON x GT and OE x GT with adjusted one-tailed side Dunnett *P*-values were used to compare the means between treatments for all variables. The frequency of metabolic disorders and infectious diseases was calculated by the FREQ procedure. The probability of occurrence of metabolic disorders and infectious diseases was calculated with the GLIMMIX procedure. The variable pre-calving silage intake rate was not normally distributed and thus was logarithmically transformed. The attributes time spent resting during pre and post-calving as well as the number of total visits to the trough, with and without feeding as well as the number of licking events, aggressions and interactions between animals were analyzed using non parametric statistics using *Wilcoxon* test as they did not follow normal distribution even after mathematical transformations. The significant differences were declared when $P < 0.05$ and a trend considered to exist if $0.05 < P < 0.10$.

## 3. Results

The interactions between treatment and days of evaluation were not significant for any of the attributes evaluated in this study. Plant extracts did not change BW and BCS in the last 21 days of the dry period (pre-calving) and in the first 21 days of lactation (post-

**Table 4**
Means for body weight and body condition score of cows fed control, oregano extract and green tea extract during the transition period.

| Variable[2] | Diets[1] | | | SEM[3] | *P* – values for Contrast | | |
|---|---|---|---|---|---|---|---|
| | CON | OE | GT | | CON x OE | CON x GT | OE x GT |
| **Body weight (Kg)** | | | | | | | |
| Pre-calving | 446.2 | 448.0 | 446.1 | 3.9 | 0.75 | 0.98 | 0.75 |
| Post-calving | 406.2 | 422.3 | 415.8 | 6.4 | 0.10 | 0.29 | 0.50 |
| Loss in the post-calving period | −9.2 | −10.8 | −17.4 | 5.5 | 0.85 | 0.32 | 0.45 |
| **BCS** | | | | | | | |
| Pre-calving | 3.50 | 3.58 | 3.50 | 0.05 | 0.38 | 0.97 | 0.38 |
| Post-calving | 3.34 | 3.34 | 3.32 | 0.06 | 0.94 | 0.81 | 0.88 |
| Loss in the post-calving period | −0.17 | −0.23 | −0.19 | 0.05 | 0.39 | 0.80 | 0.54 |

[1] Diets: CON = control; GT = 5 g / day of green tea extract; OE = 10 g / day of oregano extract.
[2] Variable: BCS = body condition score.
[3] SEM = Standard error of the mean.

**Table 5**
Means for dry matter intake and feeding behavior during the pre-calving period of cows fed control diet or diets containing oregano extract and green tea extract.

| Variable[2] | Diets[1] | | | SEM[3] | *P* – values for Contrast | | |
|---|---|---|---|---|---|---|---|
| | CON | OE | GT | | CON x OE | CON x GT | OE x GT |
| DMI (Kg) | 8.0 | 8.4 | 8.1 | 0.3 | 0.27 | 0.81 | 0.41 |
| Daily time spent Ruminating (min) | 458.4 | 518.8 | 461.0 | 25.1 | 0.11 | 0.94 | 0.12 |
| Daily time spent resting (min) | 862.2 | 885.8 | 843.2 | 42.2 | 0.82 | 0.58 | 0.44 |
| Daily time spent in activity (min) | 124.4 | 93.9 | 124.3 | 15.6 | 0.19 | 0.99 | 0.19 |
| Diurnal time spent standing up (min) | 347.3 | 378.4 | 346.5 | 14.9 | 0.15 | 0.97 | 0.16 |
| Diurnal time spent lying down (min) | 54.1 | 24.4 | 62.4 | 13.9 | 0.14 | 0.67 | 0.08 |
| Time spent at the feed trough (min) | 149.5 | 137.7 | 144.5 | 10.8 | 0.47 | 0.76 | 0.69 |
| Time spent eating the concentrate (min) | 22.7 | 20.0 | 28.6 | 2.8 | 0.51 | 0.15 | 0.05 |
| Time spent eating the silage (min) | 126.6 | 117.6 | 115.6 | 10.7 | 0.59 | 0.50 | 0.91 |
| CIR (g of DM / min) | 0.17 | 0.18 | 0.16 | 0.01 | 0.48 | 0.74 | 0.33 |
| SIR (g of DM / min) | 38.0 | 41.5 | 41.7 | 4.5 | 0.48 | 0.60 | 0.89 |

[1] Diets: CON = control; GT = 5 g / day of green tea extract; OE = 10 g / day of oregano extract.
[2] Variable: DMI = dry matter intake; CIR = concentrate intake rate; SIR = silage intake rate.
[3] SEM = Standard error of the mean.

calving) (Table 4). Extracts of oregano and green tea did not influence BW losses and BCS losses, which averaged 12.5 kg of BW and 0.20 points of BCS losses after calving (Table 4).

In the pre-calving period, DMI was not affected by treatments (Table 5). Cows in the OE group tended ($P = 0.08$) to remain less time lying down during the day and to spend less time eating the concentrate ($P = 0.05$) than cows in GT (Table 5).

After calving, OE cows tended ($P = 0.08$) to have higher DMI than CON and had a higher concentrate intake rate than GT ($P < 0.01$) and CON ($P = 0.01$) cows, spending 9.8 and 7.5 min less to eat the concentrate in relation to GT ($P = 0.01$) and CON ($P = 0.02$) cows, respectively (Table 6). Cows in the OE group tended ($P = 0.09$) to produce more milk corrected for energy (ECM) than the cows in CON while produced 26.4% more ECM than cows in GT ($P < 0.01$) (Table 6). The OE cows tended to be more efficient ($P = 0.09$), producing 0.3 kg more of milk for each kg of DM ingested compared with cows fed GT (Table 6).

During the pre-calving period, cows in OE visited less frequently the feeder (expressed as the total number of visits and visits without food ingestion) than cows in CON and GT. The cows supplemented with GT had a greater number of visits to the feeder with feed ingestion than OE ($P = 0.04$). Also GT cows tended to interact less with their group mates compared to cows in the OE treatment ($P = 0.08$) and interacted less ($P < 0.01$) compared with CON cows (Table 7). Cows fed plant extracts licked the trough less often and engaged less aggression events in relation to cows in CON (Table 7).

The occurrence of each metabolic disorder or disease as well as the total number of clinical and subclinical cases were similar across diets. Overall the first clinical symptoms or subclinical diagnosis were observed on average 8 days later (around day 12 ± 2.9 post-calving) in OE group compared with cows in GT. There were no records for placenta retention, lameness (animals with locomotion score greater than 3) and clinical hypocalcemia. The overall frequency of healthy animals (without any clinical or subclinical manifestations) was only 25% of the evaluated animals. On average, 54.2% of the animals were affected by at least one clinical disease, and 33.3% were affected by more than one disorder or disease, especially mastitis, metritis, subclinical hypocalcemia as well

**Table 6**
Means for dry matter intake and feeding behavior during post-calving cows fed control diet or diets containing oregano extract and green tea extract.

| Variable[2] | Diets[1] | | | SEM[3] | *P* – values for Contrast | | |
|---|---|---|---|---|---|---|---|
| | CON | OE | GT | | CON x OE | CON x GT | OE x GT |
| DMI (Kg) | 9.1 | 10.4 | 9.2 | 0.5 | 0.08 | 0.82 | 0.12 |
| Milk production (kg) | 16.1 | 18.6 | 13.8 | 0.9 | 0.07 | 0.06 | < 0.01 |
| ECM production (kg) | 16.7 | 19.7 | 14.5 | 1.2 | 0.09 | 0.19 | < 0.01 |
| Efficiency of milk production (kg / kg) | 1.7 | 1.8 | 1.5 | 0.1 | 0.80 | 0.11 | 0.09 |
| Efficiency of ECM (kg / kg) | 1.9 | 1.9 | 1.7 | 0.3 | 0.87 | 0.35 | 0.31 |
| Daily time spent ruminating (min) | 434.8 | 470.6 | 463.1 | 21.1 | 0.26 | 0.35 | 0.81 |
| Daily time spent resting (min) | 649.2 | 639.4 | 632.0 | 23.7 | 0.48 | 0.12 | 0.43 |
| Daily time spent in activity (min) | 356.0 | 329.9 | 344.9 | 17.9 | 0.29 | 0.64 | 0.54 |
| Diurnal time spent standing up (min) | 38.0 | 45.2 | 42.9 | 8.5 | 0.57 | 0.69 | 0.86 |
| Diurnal time spent lying down (min) | 39.7 | 72.8 | 74.4 | 23.8 | 0.36 | 0.31 | 0.97 |
| Diurnal time spent grazing (min) | 309.4 | 274.6 | 271.0 | 18.8 | 0.22 | 0.16 | 0.90 |
| Time spent eating the concentrate (min) | 27.3 | 19.8 | 29.4 | 2.1 | 0.02 | 0.49 | 0.01 |
| CIR (g of DM / min) | 0.17 | 0.24 | 0.14 | 0.02 | 0.01 | 0.29 | < 0.01 |

[1] Diets: CON = control; GT = 5 g / day of green tea extract; OE = 10 g / day of oregano extract.
[2] Variable: DMI = dry matter intake; ECM = energy corrected milk; CIR = concentrate intake rate.
[3] SEM = Standard error of the mean.

**Table 7**
Means for social behavior during pre-calving cows fed a control diet or diets containing oregano extract and green tea extract.

| Variable | Diets[1] | | | SEM[2] | P – values for Contrast | | |
|---|---|---|---|---|---|---|---|
| | CON | OE | GT | | CON x OE | CON x GT | OE x GT |
| Visits to the feed trough (n°) | 13.9 | 10.4 | 16.2 | 2.2 | < 0.01 | 0.57 | < 0.01 |
| Visits to the feed trough with food ingestion (n°) | 7.9 | 7.0 | 10.3 | 1.9 | 0.42 | 0.16 | 0.04 |
| Visits without food ingestion (n°) | 6.0 | 3.4 | 5.9 | 1.3 | < 0.01 | 0.50 | 0.01 |
| Duration of each visits to the trough with food ingestion (min) | 20.9 | 19.8 | 16.2 | 4.3 | 0.86 | 0.45 | 0.59 |
| Licking the trough (n°) | 3.5 | 1.2 | 1.4 | 1.0 | 0.02 | 0.03 | 0.87 |
| Social interactions (n°) | 2.5 | 2.1 | 0.9 | 0.5 | 0.32 | < 0.01 | 0.08 |
| Aggression (n°) | 5.2 | 2.7 | 2.8 | 0.8 | < 0.01 | 0.04 | 0.27 |

[1] Diets: CON = control; GT = 5 g / day of green tea extract; OE = 10 g / day of oregano extract.
[2] SEM = Standard error of the mean.

as clinical and subclinical ketosis (Fig. 1).

## 4. Discussion

The present study was designed to evaluate the effects on intake, behavior and health status of Jersey cows fed with oregano extract or green tea during the transition period. Oregano and green tea extracts have been tested in dairy cows primarily for their potential to modulate rumen metabolism and methane emissions (Hristov et al., 2013; Kolling et al., 2018), but little is known about the effects of these extracts on behavioral and immune responses (Winkler et al., 2015; Kolling et al., 2016) especially during the transition period. In the present study, we highlighted the tendency for positive effects of supplying oregano extract on the DMI in the post-calving, as well as changes in feeding and social behavior when oregano and green tea extracts are supplied to dairy cows in the transition period, and this is the main contribution of the present study.

After calving, the energy demand for maintenance and lactation usually exceeds the amount consumed, predisposing high-yielding dairy cows to negative energy balance (Bauman and Currie, 1980), with mobilization of the body tissues. Plant extracts did not help to decrease body tissue mobilization during the transition period as losses in BW and BCS were similar to CON, but the tendency of higher DM intake and the significant higher concentrate intake rate observed in OE cows during the first weeks post-calving could have beneficial effects. Increased intake minimized the negative energy balance contributing to prevent health and reproductive problems (Trevisi et al., 2012; Drackey and Cardoso, 2014), but the present study did not evidenced it.

Oregano might have increased the concentrate intake rate due to its action on the feeding center in the central nervous system (Simansky, 1995; Trabace et al., 2011). Oregano contains carvacrol (Lagouri et al., 1993) that may decrease the 5-HT receptors in the central nervous system by reducing the concentration of the neurotransmitter serotonin (Trabace et al., 2011), stimulating the feeding activity (Simansky, 1995). This ability of carvacrol to modulate the central nervous system may explain the tendency of increased DMI after calving and the greatest concentrate intake rate, although the concentration of 5-HT receptors and serotonin

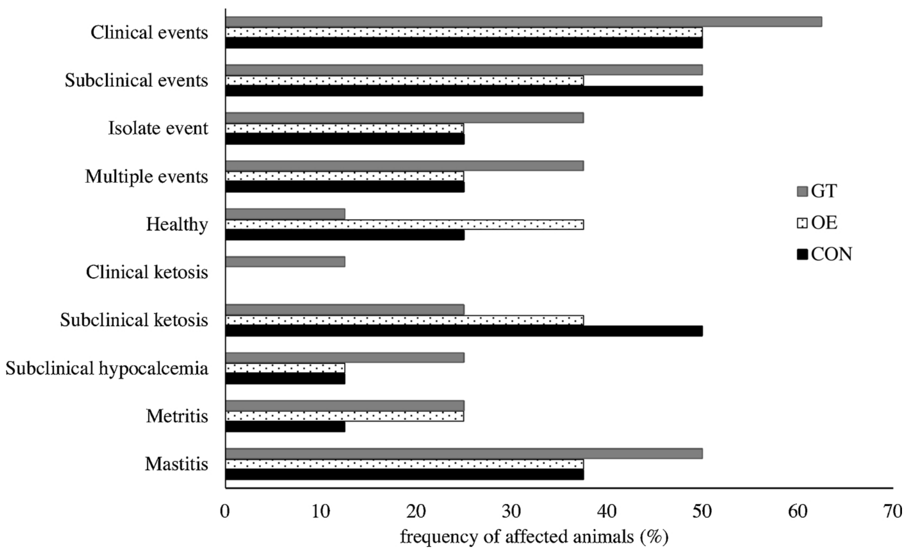


**Fig. 1.** Frequency (%) of Jersey cows affected by metabolic disorders and infectious diseases fed diets without (CON) or with green tea (GT) and oregano extracts (OE) during the transition period.

neurotransmitter were not measured in this study.

In the literature, the effects of the essential oils on DMI are still contradictory. Gabbi et al. (2009a) observed faster concentrate intake for dairy heifers supplemented with 1 g/day of a blend of essential oils. On the other hand, Lejonklev et al. (2016) and Kolling et al. (2018) did not observe modifications in the DMI of lactating cows with the inclusion of 0.2 and 1 g / kg of DM of oregano oil and 10 g / day of oregano extract into the diets, respectively.

The above trend in DM intake and positive effect on concentrate intake rate resulted in the tendency of higher milk and ECM production in OE cows compared with the control and in higher milk and ECM production in OE cows compared with GT. The lack of significant effect of oregano extract on feed efficiency is in agreement with other studies that have demonstrated reduced methane emissions without changes in feed efficiency (Kolling et al., 2018).

The rumination process is a key component of digestion and ingestion (Gregorini et al., 2013), depending mainly on the composition of the diet, especially the effective fiber content (Schirmann et al., 2012). In this study, cows ruminated on average 8.0 and 7.6 h per day in the pre and post-calving, respectively (Tables 5 and 6), in agreement with Adin et al. (2009). On the other hand, the differences in DMI were small and the basal diet composition was the same for all cows what explain the similar rumination time between the treatments.

Both oregano and green tea extracts contain chemical compounds that may interact with the central nervous system (Mirza et al., 2013; Zotti et al., 2013) triggering changes in cognitive and emotional processes, eliciting feelings of well being (Mirza et al., 2013). In rats, carvacrol increased immobility (Trabace et al., 2011), while catechins reduced anxiety and stimulated locomotion and exploration of the environment (Mirza et al., 2013). The direct effect of the extracts on the central nervous system was not measured in the current study, but we hypothesized that it explains the reduction in the aggressive behavior and licking events observed in GT and OE groups compared with CON as well with the differences in the number of visits to the trough observed for OE and GT cows, as OE decreased the number of visits to the through during pre-calving compared to GT. The reduction in aggressive interaction in GT compared with CON might be related to reduced anxiety effects usually attributed to green tea.

During the transition period dairy cows face several physiological and metabolic changes such as reduction in circulating levels of glucose and insulin (Sordillo and Raphael, 2013), increase in growth hormone concentration with decoupling of the somatotropic axis (Leroy et al., 2008) as well as increase concentrations of glucocorticoids and catecholamines (Weber et al., 2001). All these changes occur in order to support pregnancy and then ensure the initiation of lactation, and affect the immune system, predisposing animals to the occurrence of diseases (Sordillo and Raphael, 2013). One of the hypotheses of the present study, that oregano and green tea extracts would improve the health state of the animals was not confirmed, as the occurrence of diseases and disorders did not differed between treatments. We are aware of some limitations of our study as we used a small number of animals, with high coefficient of variation that may have prevented the detection of significant differences between the treatments. The transition period is a very challenging phase for animal health and perhaps higher doses could have reduced the occurrence of diseases and metabolic disturbances.

## 5. Conclusions

Oregano extract but not green tea extract tended to present beneficial effects on DMI and feeding rate in Jersey dairy cows during the first weeks after calving. Extracts influenced distinctly feeding and social behavior, such as the number of visits to the trough, social and aggressive interactions. Cows fed oregano extract tend to produce more milk and to be more efficient than cows fed green tea extract. Plant extracts did not reduce the occurrence of diseases and metabolic disorders. Considering the improvements in intake rate, feed efficiency and feeding and social behavior traits, oregano extract may contribute to enhance welfare during the transition period.

## Declaration of Competing Interest

None.

## Acknowledgements

The authors thank to Brazilian National Research Council - National Council for Scientific and Technological Development 473562 / 2012-0 for the research and fellow research grants, to the Brazilian Agricultural Research Corporation - EMBRAPA Temperate center for providing the animals, structure and employees for the experiment, Higher Education Personnel Improvement Coordination for fellow research grants and to Mr. Ivan dos Santos for the supply of the oregano extract.

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