Faculty of Science

Effect of dietary Oregano supplementation on growth performance and lipid oxidation stabilty muscle: Guinea pigs as a meat animal model.

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Outline of Presentation

- ***General Background***
- ***Objective/Hypothesis***
- ***Experimental approach***
- ***Results and Discussion***
- ***Conclusion***

Background

- Plant Essential Oils (EO) have proven to be effective as antimicrobial, antioxidant and anti inflammatory agents *in vitro.*

- *Some in vivo* data suggests potential use of EOs as growth promoters in animal diets (Cardozo et al.,2004;Ouwehand et al.,2010)

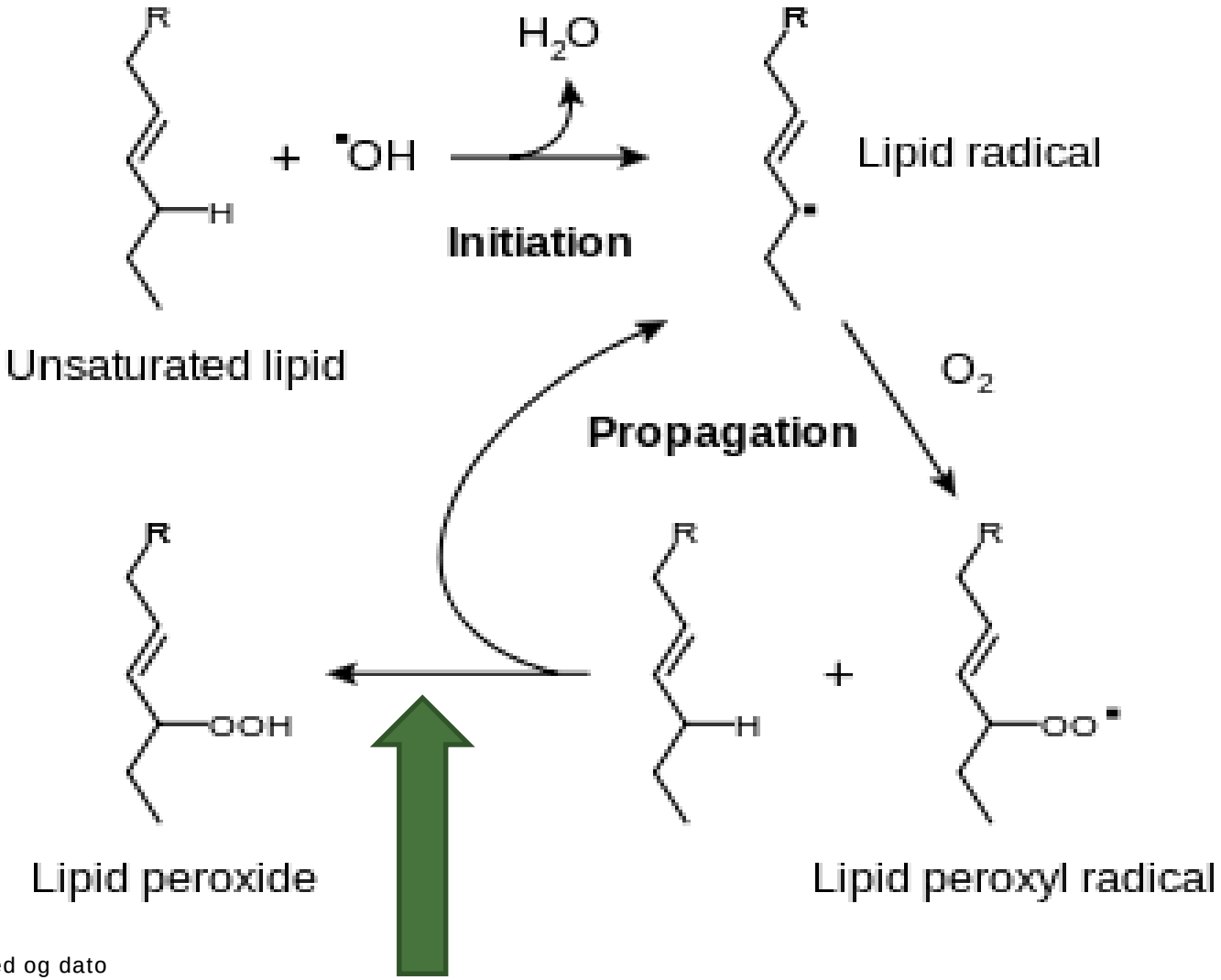
- Oregano is reported as one of such EO with high antimicrobial, antioxidant, antifungal effects. (Luna et al., 2010;Avila-Ramos et al., 2012)

- **Oregano** is an aromatic medicinal plant especially found in the Mediterranean area.
- Activity of the oregano essential oil is mainly distributed through the main components **CARVACROL** and **THYMOL**.
- This substances have bioactivity on physiology and metabolism of the animal(Reiner et al.,2009).
- Modify the bacterial cell membrane permeability (Lambert et al., 2001) and reacts with lipid hydroxyl radicals converting them into stable products (Yanishlieva et al., 2001).

Summary of Oregano EO effect on Poultry and Pigs

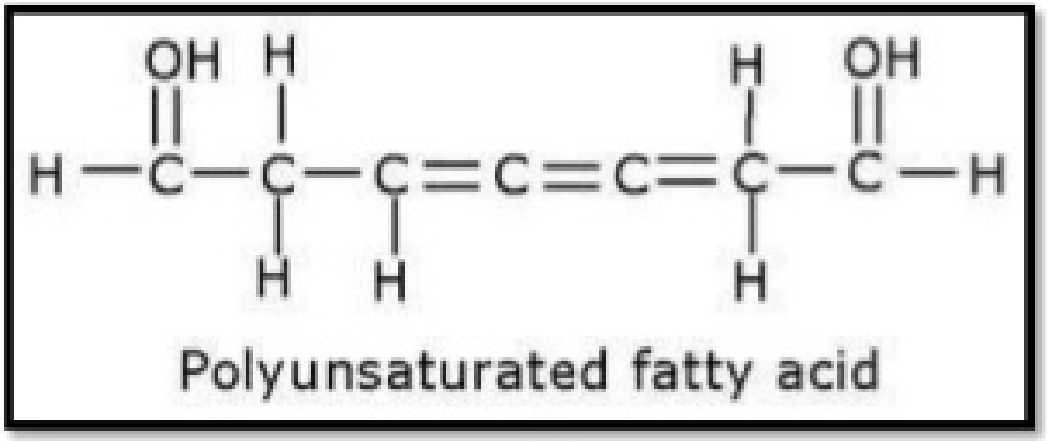
Species	Report	Reference
Turkey	Delayed lipid oxidation in fresh and cooked meat	Botsoglou et al., 2003b
Rabbits	Oregano EO improved growth performance, delays lipid oxidation and control microbial population	Soultos et al., 2009 Botsoglou et al., 2004
Chickens	Delay lipid oxidation and no effect on growth performance	Botsoglou et al., 2003a
Pigs	Lipid Oxidation was not delayed in pork	Simitzis et al., 2010 Janz et al., 2007
Pigs	Improved intake, low mortality, increased performance	Allan and Bilkei, 2005

Lipid Oxidation Cycle



Factors Causing Rancidity

- Catalysts: trace metal ions & inorganic salts
- Temperature
- Amount of PUFA
- Time
- Light
- Water



Polyunsaturated fatty acid

Objectives of the Study

1. To study the effect of dietary Oregano on growth performance by measuring feed intake, weight gain and feed conversion efficiency

2. To study the antioxidant effect of the dietary Oregano on lipid oxidation stability of muscles muscle stored over time

Hypothesis

Dietary oregano will improve growth performance of guinea pigs and delay lipid oxidation in meat

Specific Objective 1

To study the effect of dietary Oregano on growth performance by measuring feed intake, weight gain and feed conversion efficiency

Experimental Design

- Total of 15 one week old guinea pigs were divided into 3 treatments (n=5).

- **Control group**: Basal Diet only
- **Linseed group**: Basal diet + 50ml linseed oil
- **Oregano group** : Basal diet + 50ml linseed oil + 1g/kg oregano

- Feeding lasted for **49 days**

- Feed intake, weight gain was recorded weekly and before euthanization

$$FCE = \frac{Total\ Feed\ Intake}{Total\ Weight\ gain} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (1)$$

$$ADG\ \left(\frac{g}{day}\right) = Total\ weight\ gain\ /age \ldots\ldots\ldots\ldots\ldots\ldots (2)$$

- Minitab statistical software was used in the statistical analysis
- Analysis was conducted as a repeated measures in a general linear model (GLM) ANOVA
- Probability value set at $p \leq 0.05$.

Results

Table 1 : Results on growth performance parameters

Performance Parameters	Control	Linseed Oil	Oregano
Initial Weight (g)	140.0 ± 15.1$_a$	136.08 ± 9.6$_a$	142.8 ± 7.4$_a$
Final Weight (g)	531.7 ± 27.1$_b$	546.2 ± 33.5$_b$	555.3 ± 32.8$_b$
ADG(g)	7.79 ± 0.36$_c$	8.37 ± 0.59$_c$	8.39 ± 0.64$_c$
ADFI(g)	33.40 ± 0.27$_d$	31.47 ± 0.48$_d$	26.41 ± 0.24$_e$
FCE	4.28 + 0.75$_f$	3.76 + 0.81$_f$	3.14 + 0.38$_h$

$_{a-h}$ Means with the same subscript in the same row are not significantly different. (n=5)

Discussion

- No significant difference intake between Linseed and Control treatments.

- contrary to the expectation that high energy of the Linseed oil will cause a significant reduction in intake for Linseed and Oregano groups in relation to the Control group.

- No significant effects of oregano on performance parameters such as feed intake, weight gain and FCE in rabbit (Botsoglou *et al.* 2004)

Discussion

- Intake was significantly reduced in Oregano supplementation resulting in a better FCE
- ADG was not significantly different in all treatments
- Dietary oregano leaves did not improve growth performance in Lambs and similarly reported in broilers (Botsoglou et al. 2004; Bampidis *et al.* 2005)
- However the FCE was significantly different in the treatments, Oregano group had a better FCE than the Linseed group and control groups.

Specific Objective 2

To study the antioxidant effect of the Oregano on lipid oxidation of guinea pig loin muscle stored over time

Method

- Thiobarbituric acid reactive substances - **TBARS** - are formed as a by-product of lipid oxidation which can be detected by the TBARS assay.

- Assay of TBARS measures malondialdehyde (MDA) present in the sample, and also as MDA from lipid hydro peroxides by the hydrolytic conditions of the reaction.

- Longissimus dorsi muscle was sampled from all pigs in each treatment group.

Methods

- A subsample from each animal was stored at -80^0 C until fatty acid composition analysis
- 2.5g of each sample for each storage day was used in the TBARS analysis and this was done in triplicates.
- FA analysis was also conducted on each animal in all treatment group in triplicates
- Extraction of fat in the samples by the method described by Segura & Lopez-Bote (2014) and later set up in the Gas Chromatograph.

- TBARS was measured using the equation below

$$\textbf{TBARS [µmol/kg]} = \frac{(\text{Abs} - \beta)}{\alpha\ [\mu\text{mol/1}]} \times \frac{(((\%H_2O / 100) * g) + 15) * 1000\ [1])}{g * 1000\ [kg]}$$

- Amount of each Fatty acid component was measured using the area of the FA on the chromatograph peak expressed as a percentage of the total fatty acid components.

- Fatty acid composition is measured as (g/100g of total fatty acids).

Results

Table 2 TBARS (µmol/kg) on dry weight value (Means ± SD) against the storage time

	Storage Time (Days)			
Treatments	**0**	**3**	**7**	**10**
Control	0.14 ± 0.00^{e}	1.08 ± 0.12^{d}	2.86 ± 0.28^{b}	4.22 ± 0.18^{a}
Linseed	0.12 ± 0.014^{e}	1.80 ± 0.12^{cd}	4.31 ± 0.09^{a}	5.12 ± 0.42^{a}
Oregano	0.10 ± 0.00^{e}	1.37 ± 0.03^{d}	1.51 ± 0.05^{d}	2.86 ± 0.06^{bc}

$^{a\text{-}e}$ Means with same superscripts all across the table are not significantly different: ($p<0.001$)

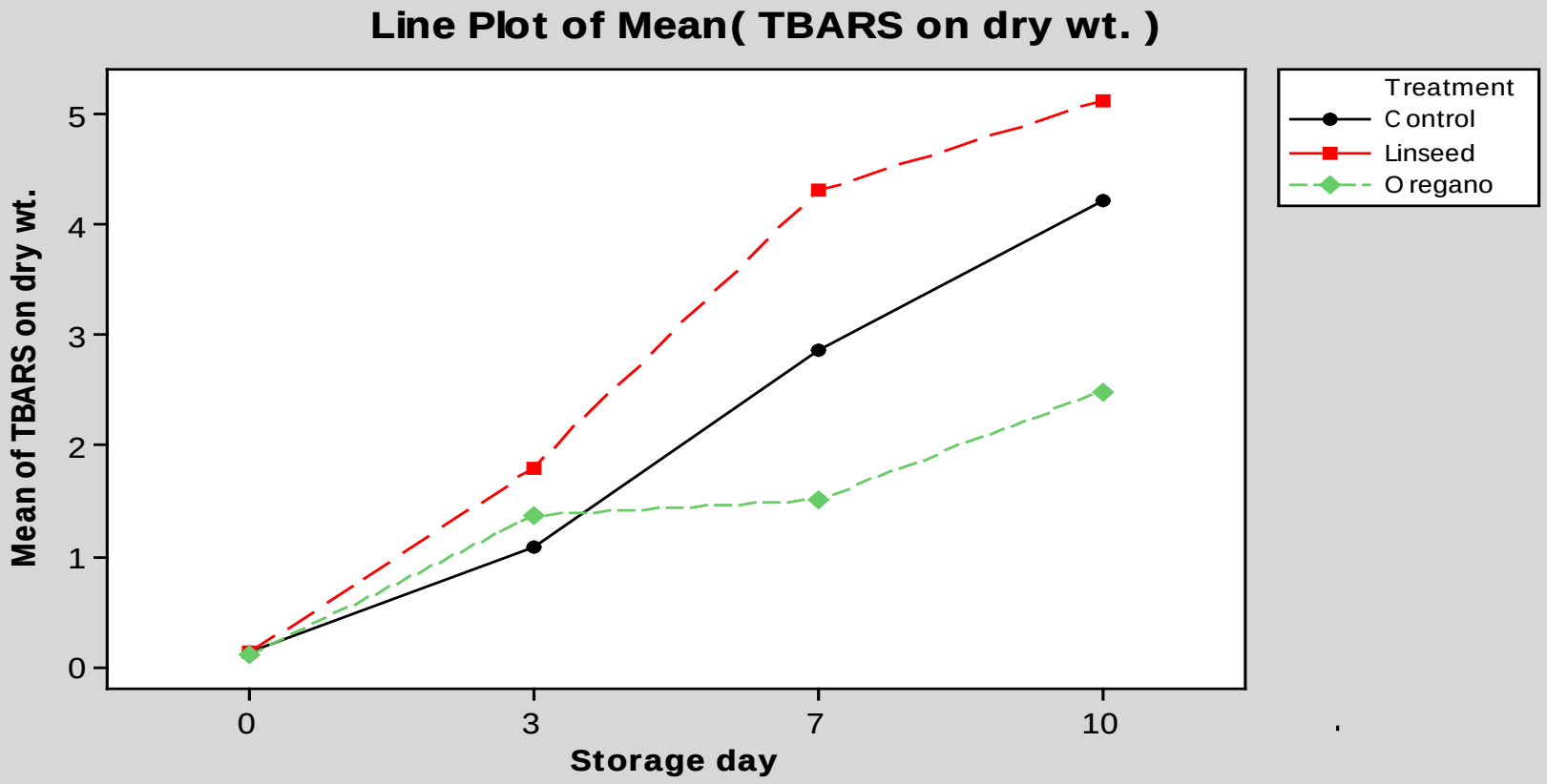


Figure 1 Effects of dietary oregano supplementation as a function of storage on lipid oxidation in guinea pig muscle as compared to a Control and Linseed oil group.

Table 3 Fatty acid composition (g/100g of total FAs: Mean + SD) in the longissimus dorsi muscle of guinea pigs fed three experimental diets

Fatty Acid	Control	Linseed	Oregano	*P- value*
C14:0	1.01 ± 0.09^{a}	0.59 ± 0.03^{b}	0.72 ± 0.22^{b}	0.000*
C16:0	23.5 ± 0.23^{c}	15.1 ± 1.94^{d}	17.4 ± 0.48^{d}	0.000*
C17:0	0.68 ± 0.55^{e}	0.56 ± 0.03^{f}	0.45 ± 0.02^{g}	0.001*
C18:0	10.9 ± 1.71^{h}	11.3 ± 1.78^{h}	7.9 ± 0.54^{h}	0.053
C18:1	23.1 ± 1.11^{j}	20.2 ± 0.23^{k}	21.9 ± 0.15^{j}	0.005*
C18:2	35.8 ± 0.22^{m}	34.4 ± 1.14^{m}	31.4 ± 0.44^{n}	0.001*
C18:3	4.98 ± 0.28^{p}	17.94 ± 0.67^{q}	19.75 ± 0.72^{r}	0.000*
C20:4	0.00 ± 0.00^{s}	0.094 ± 0.16^{s}	0.188 ± 0.163^{s}	0.295 NS
C20:5	0.00 ± 0.00^{t}	0.31 ± 0.30^{t}	0.152 ± 0.13^{t}	0.218 NS
C22:6	0.00 ± 0.00^{u}	0.08 ± 0.13^{u}	0.12 ± 0.10^{u}	0.383 NS
PUFA	40.61 ± 0.15^{v}	53.48 ± 0.49^{w}	52.05 ± 0.280^{x}	0.000*
n6: n3	7.145^{y}	1.865^{a}	1.570^{z}	0.000*

NS: Not significant;
**Significant*
PUFA: Total polyunsaturated fatty acid
n6=Omega 6 fatty acid
n3=Omega 3 fatty acid
$^{a\text{-}z}$ *Means within the same row having same superscripts are not significantly different*

Discussion

- TBARS values between control and oregano treatment suggests the effect of oregano in delaying lipid oxidation
- consistent with reports in chickens, turkeys and rabbits , in delaying lipid oxidation (Botsoglou et al., 2003); papageorgiou et al., 2003)
- Although the Oregano group had a higher PUFA, than the control group, the level of TBARS was significantly lower.
- Data from the Oregano group show a lower n6/n3 ratio, which is an indication of a healthy fatty acid componsition of the meat.

Conclusion

Results suggest the effects of the Oregano in delaying lipid oxidation and improving growth performance by increasing feed efficiency

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Thank You

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SLU
Swedish University of Agricultural Sciences