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# Tolerance and efficacy study of palygorskite incorporation in the diet of laying hens



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#### A R T I C L E I N F O

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#### ABSTRACT

The objective of this study was to assess the safety and effectiveness of palygorskite inclusion in the diet of laying hens. A total of 135 Lohmann Brown hens, aged 21 weeks, were randomly assigned to one of three treatment groups (n = 45): group C (control), group S (fed a diet with 1% palygorskite, standard-level) and group T (fed a diet with 9.1% palygorskite, multi-fold of standard-level). All hens were kept in commercial laying cages (3 hens per cage, 15 replicates per group). Hens of group T were used only for the first eight weeks (tolerance study period), while hens of groups C and S were kept for another sixteen weeks (efficacy study period). In each group, laying percentage, feed intake, feed conversion ratio, mortality and egg quality characteristics were assessed. The results showed that feeding a ration with 9.1% palygorskite (group T) did not have any negative effects on laying percentage (%), feed conversion ratio or health of hens. However, the body weight of hens in group T was significantly lower (P < 0.001) while their feed intake was higher (P < 0.001) compared to those of groups C and S for the 8-week experimental period. Furthermore, the produced eggs had significantly (P < 0.001) lighter coloured egg yolk. The incorporation of 1% palygorskite in the ration for 24 weeks improved laying percentage (by 2.6%, P < 0.05) and feed conversion ratio (by 4.6%, P < 0.05); feed intake and body weight of hens were unaffected. The produced eggs had significantly lighter eggshell and yolk colour (P < 0.001) as well as increased eggshell thickness (P < 0.05). It was concluded that palygorskite is a tolerable material which has the potential to improve laying hen performance and influence egg quality traits when used at recommended levels. © 2014 Elsevier B.V. All rights reserved.

#### 1. Introduction

Intensive research, carried out in recent decades, has confirmed the benefits of clay mineral inclusion in animal nutrition. Studies report that such materials can be used as technological additives in order to improve feed manufacture (Angulo et al., 1995, 1996), as absorbents for mycotoxins (Phillips, 1999; Huwig et al., 2001), or as enhancers of the nutritive value of animal diets (Parisini et al., 1999; Ouhida et al., 2000).

A magnesium–aluminum silicate mineral that has attracted increasing interest in recent years is palygorskite (attapulgite). Although the term palygorskite is the official name assigned by the International Nomenclature Committee (Murray, 1999), in practice, the word attapulgite is widely used by producers and users.

Palygorskite is widely known as an active ingredient in pharmaceutical applications. Its high sorption capacity and large specific surface area account for its use as gastrointestinal protector and as antidiarrheic. It can also act as antacid, since adsorption of H<sup>+</sup> ions to the

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mineral surface induces gastric acidity reduction (Carretero and Pozo, 2010).

However, due to its absorptive, rheological and catalytic properties, palygorskite is beneficial for many other industrial uses as well as applicable in animal nutrition (Murray, 2000). It finds extensive usage either as a binding agent for the pelletization of animal feed or as an animal feed supplement. In 2005, palygorskite entered for the first time the Community Register of Feed Additives being listed under silage additive functional group. However, it should be noted that since 2011 palygorskite has been included in the catalogue of feed materials (Commission regulation 575/2011).

Nevertheless, in comparison with other clays, in vivo studies testing inclusion of palygorskite in animal diets are sporadic. The efficacy of palygorskite in counteracting the deleterious effects of aflatoxin contaminated diets on performance and serum profiles of weanling pigs was tested by Schell et al. (1993). Its dietary inclusion improved average daily gain and average daily feed intake while it prevented increased serum ALP and GGT levels compared with those of pigs fed the aflatoxin control diet. Papas et al. (2010) reported that addition of 1%m/m palygorskite in broiler diets improved the hardness of pellets suggesting, therefore, its application as an agglomerant. Inclusion of palygorskite in weaned piglet diets exerted a protective effect against diarrheal infections leading to a concomitant improvement in their

 $<sup>\</sup>ast\,$  Corresponding author at: P.O. Box 393, GR-54124, Thessaloniki, Greece. Tel.: +30 2310999808; fax:  $+\,30$  2310999892.

growth performance (Zhang et al., 2013). Furthermore, dietary palygorskite supplementation, at levels up to 10 kg/ton, in lactating cow diets increased milk protein yield and decreased milk colony forming units, rendering other productions traits unaffected (Bampidis et al., 2014).

However, information of its application in laying hen rations is lacking. For this reason, our interest was drawn to designating a study that would assess the safety and the effectiveness of palygorskite inclusion in the diet of laying hens.

#### 2. Materials and methods

#### 2.1. Birds and management

A total of 135 Lohmann Brown hens, aged 21 weeks, were randomly allocated into three treatment groups: group C (control), group S (palygorskite used at a use-level) and group T (palygorskite used at a multi-fold of the use-level). Each treatment group consisted of 45 hens placed in fifteen 3-deck cages (3 hens per cage). Each cage was equipped with designated feeders and drinking nipples to ensure similar feeding and drinking space across groups.

#### 2.2. Feeds and feeding

All hens were offered free and continuous access to a nutritionally non-limiting concentrate diet designated as basal diet for group C, whereas in group S and group T the hens were offered the same diet supplemented with palygorskite at a level of 1% and 10% of the quantity of basal diet, respectively. So the level of the tested material in the diets C, S and T was 0%, 1% and 9.1% respectively. The physical and chemical compositions of the diets are shown in Table 1.

#### Table 1

Physical composition and chemical analysis of the experimental diets.

	Group						
	С		S		Т		
	kg	%	kg	%	kg	%	
Physical composition							
Corn (flour)	531	53.1	531	52.6	531	48.3	
Soybean meal 44%	295	29.5	295	29.2	295	26.8	
Calcium carbonate	80	8	80	7.9	80	7.3	
Wheat bran	40	4	40	4.0	40	3.6	
Soya oil	28	2.8	28	2.8	28	2.5	
Dicalcium phosphate	15	1.5	15	1.5	15	1.4	
Mineral & vitamin premix <sup>a</sup>	10	1	10	1.0	10	0.9	
Methionine	0.5	0.05	0.5	0.0	0.5	0.0	
Sodium carbonate	0.5	0.05	0.5	0.0	0.5	0.0	
Palygorskite	-	-	10	1.0	100	9.1	
Total	1000	100	1010	100	1100	100	
Chemical composition <sup>b</sup>							
Crude protein, %	17.62		17.44		16.02		
Crude fibre %	3.66		3.62		3.33		
Fat, %	5.44		5.39		4.95		
ME, Kcal/kg	2767.8		2740.4		2516.2		
Ca, %	3.35		3.32		3.05		
Phosphorus, %	0.46		0.45		0.41		
Lysine, %	0.98		0.98		0.89		
Methionine, %	0.34		0.34		0.31		

<sup>a</sup> Providing per kg of diet: 12,500 IU vitamin A, 3000 IU vitamin D3, 30 mg vitamin E, 3 mg vitamin K3, 2 mg vitamin B1, 5 mg vitamin B2, 3 mg vitamin B6, 0.02 mg vitamin B12, 30 mg nicotinic acid, 12 mg pantothenic calcium, 1 mg folic acid, 0.05 mg biotin, 300 mg vitamin C, 20 mg Car. red, 500 mg choline chloride, 0.25 mg Co (carbonate) E3, 2 mg iodine (potassium iodide) E2, 0.15 mg Se (sodium selenate) E8, 60 mg Fe (sulphurated) E1, 150 mg Mn (MnO) E5, 12 mg Cu (oxide) E4, 60 mg Zn (ZnO) E6, 10 mg antioxidant 5×.

<sup>b</sup> Calculated from feeding tables.

#### 2.3. Palygorskite material

The palygorskite used in the present study (AFG-60 mesh) was provided by Geohellas S.A., Greece. It was mined from deposits, located in Ventzia basin of Grevena, western Macedonia, Greece.

Palygorskite is a magnesium–aluminum silicate mineral that along with sepiolite compose the group of hormites (Harvey and Murray, 1997). It consists of double silica tetrahedral chains linked together by octahedral oxygen and hydroxyl groups containing Al and Mg ions in a chain-like inverted structure. The inverted tetrahedra occur regularly and cause channels through the structure. These channels together with the elongate habit and the fine particle size, give palygorskite a high surface area. Furthermore, the charge on the particles, the channels through the structure and the high surface area, provide this clay mineral with a high absorption and adsorption capacity while the elongate particles induce higher viscosity when palygorskite is added to any liquid (Murray, 1999).

The mineralogical characterisation of the material used in our study comprised a minimum of 75% palygorskite and 25% bentonite–saponite. The chemical composition and some properties of this material are given in Table 2.

#### 2.4. Experimental design

All experimental work was approved by the Veterinary Directorate of Thessaloniki (Decision 13/14557 12-11-2008) as required by national and EU legislation. The experimental design was in accordance with the standards and guidelines of the European Food Safety Authority (EFSA, 2008). The tolerance study was conducted to provide evidence for the safety of palygorskite inclusion in the laying hen diet when consumed at higher than recommended doses. The design included three groups: a control group (C), a use level group (S) and a tolerance group with the multi-fold of the use level (T). The hens used were 21 weeks old at the beginning of the experiment as they had to be in the first third of the laying period during the trial. The duration of the tolerance study was eight weeks. At the end of this period layers of group T were removed while the experiment continued with hens of groups C and S for another sixteen weeks in order to estimate the effectiveness of palygorskite for a total period of twenty-four weeks (efficacy study).

#### 2.5. Measurements

#### 2.5.1. Performance traits

All hens were individually weighed at the start of the experiment, as well as at the end of the 8th and 24th weeks. Egg production was recorded daily while at the same time any dead birds were counted. Laying performance was expressed as % laying percentage that was based on the existing number of hens at the time of measurement. Amounts of feed provided to hens in each cage were written down every day. On a weekly basis feed refusals in individual cages were measured in order to calculate the daily feed intake and feed conversion ratio (kg feed/kg eggs produced).

Table 2	2
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Chemical composition and properties of the palygorskite clay.

Chemical composition (wt.%)	Properties	
MgO 15.05 Al <sub>2</sub> O <sub>3</sub> 4.97	Apparent bulk density (g/cm <sup>3</sup> ) Specific surface area (m <sup>2</sup> /g) Water absorption (wt.%—Westinghouse method) pH (5% dispersion)	0.65 150 150–200 8.5

<sup>a</sup> Loss on ignition.

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