g-pur.com

G-SCIENCE

5500 Highlands Parkway
Smyrna, GA 30082
USA
Tel. +1 678 306 2505
Fax +1 678 306 2500

David Edwards, Ph.D.
Director
Division of Animal Feeds, HFV-220
Center for Veterinary Medicine
Food and Drug Administration
7519 Standish Place
Rockville, MD 20855

October 17, 2018

Subject: **GRAS Notice Filing**
Clinoptilolite as an Anticaking Agent

Dear Dr. Edwards

We respectfully resubmit the attached Notice in support of the determination that clinoptilolite, a natural crystalline aluminosilicate mineral, is Generally Recognized as Safe (GRAS) when used as an anticaking agent in animal feed for major food animals (cattle, swine, chicken, turkeys), other minor species (ruminants: goats and sheep, other poultry species) and pet animals (cats, dogs) at levels up to 1%.

G-Science has determined that clinoptilolite produced by Zeocem a.s. is GRAS based on scientific procedures in accordance with 21 CFR 570.30(a) and (b). The enclosed GRAS Notice provides a review of the information related to the intended uses, manufacturing, and safety of the ingredient. The GRAS notice was compiled in accordance with the rules and regulations set out in the Federal Register, Vol. 81, No. 159, Part 570, Subpart E. The analytical data, published studies, and information that are the basis for this GRAS determination are included with this Notice. The GRAS Notice consists of seven parts as required by 21 CFR 570.225 through 570.255. We have included one hard copy of the GRAS Notice and its Attachments.

Certain data and information included in some sections of this notice are exempt from disclosure under the Freedom of Information Act, 5 U.S.C 552 due to being considered a trade secret and/or commercial information that is confidential. Those sections and information/data contained therein which meet these criteria are identified using a "grey" background. They are described in Section 1, part (c)(8). If you disagree with our

g-pur.com

G-SCIENCE

5500 Highlands Parkway
Smyrna, GA 30082
USA
Tel. +1 678 306 2505
Fax +1 678 306 2500

confidential aims, we respectfully request that the Agency notify us, prior to any release of the Notice on FDA´s website or otherwise.

We trust that this resubmission satisfies the Agency´s needs and addresses all deficiencies that were identified during discussion of the first submission process in 2017, and will be deemed accepted and complete. Should any questions arise, please contact us, preferably by telephone (+1 678 925 8015) or e-mail (thomas.berger@g-science.com) so that we can promptly reply.

Sincerely yours,

Thomas Berger
Position: Vice President G-Science, Inc.

RECEIVED
OCT 24 2018
By

Enclosures

Inhaltsverzeichnis | contents

TAB

PART 1

Generally Recognized as Safe (GRAS) Notice

for

Clinoptilolite
(CAS Reg. No. 12173-10-3)

Submitted to:

Division of Animal Feeds (HFV-220)
Office of Surveillance and Compliance
Center for Veterinary Medicine
Food and Drug Administration
7519 Standish Place
Rockville, MD 20855

Notifier:

G-SCIENCE, Inc.
5500 Highlands Parkway
Smyrna, GA, 30082
USA

Part 1

SIGNED STATEMENTS AND CERTIFICATIONS

Table of Contents

1. SIGNED STATEMENTS AND CERTIFICATIONS

1.1 Claim of GRAS Notice

G-SCIENCE, Inc. (hereinafter G-Science or the Notifier) submits the enclosed notice to support the claim that the compound clinoptilolite, a natural crystalline aluminosilicate mineral of sedimentary origin, is generally recognized as safe (GRAS) when used as an anticaking agent in feed products for livestock (ruminants and swine), poultry and in pet food (dogs and cats).

The enclosed notice is a resubmission of the previous notice AGRN #25 submitted on August 21, 2017 for which evaluation was ceased on our request on May 4, 2018. This resubmitted notice consolidates information from the original notice and an amendment prepared by the Notifier in response to questions raised by FDA/CVM during telephone conferences and one meeting.

1.2 Name and Address of the Notifier

G-SCIENCE, Inc.
5500 Highlands Parkway
Smyrna, GA, 30082
USA
Phone: +1 678 925 8015

1.2.1 Person Responsible for the Submission

Thomas Berger
Vice President G-SCIENCE, Inc.
Email: thomas.berger@g-science.com

1.3 Name of the Notified Substance

The name of the notified substance is clinoptilolite, a natural crystalline aluminosilicate mineral of sedimentary origin. The Chemical Abstracts Service (CAS) identifies the substance using the registry number 12173-10-3 (CAS, 2016).

1.4 Intended Conditions of Use

Clinoptilolite is intended to be used as an anticaking agent. It is added to complete feed mixtures or feed premixes of diets of livestock (ruminants, swine), poultry and pets, regardless of stage of production, at a level not exceeding 1% (10,000 mg/kg feed) by weight of the feed composition in accordance with good manufacturing or feeding practice.

1.5 Basis for Conclusion of GRAS Status

The determination of the GRAS status of clinoptilolite is based on scientific procedures in accordance with the Code of Federal Regulations (CFR) 21 CFR 570.30(a) and (b).

1.6 GRAS Conclusion

The Notifier claims that because the notified substance is GRAS the substance is exempt from the requirement for premarket approval as defined in Chapter III, Sec. 301 [21 U.S.C. 331] (II)(3)(C) of the Federal Food and Drug Cosmetic Act [As Amended Through P.L. 114-146, Enacted April 19, 2016] (OLRC, 2016).

The GRAS notice was compiled in accordance with the rules and regulations set out in the Federal Register, Vol. 81, No. 159, Part 570, Subpart E (GPO, 2016a). The Notifier certifies to the best of its knowledge that the GRAS notice is a complete, representative and balanced submission that includes unfavorable information, as well as favorable information, that is known to the Notifier, and pertinent to the evaluation of the safety and GRAS status of the use of the substance

1.7 Availability of Information

The GRAS notice is being submitted in electronic format. The Notifier will retain copies of all data and information that form the basis for the Notifier's conclusion of GRAS status. The Notifier agrees to provide FDA, either during or after its evaluation of the notice, complete copies of the data and information, either in electronic format accessible for evaluation or on paper. FDA can review or copy the data and information during customary business hours at the Notifier's address given above. Requests for copies of the respective materials may be directed to the person responsible for the submission.

1.8 Exemption from Disclosure under the FOI provisions

Certain data and information included in some sections of the notice are exempt from disclosure under the Freedom of Information Act, 5 U.S.C 552 due to being considered a trade secret and/or commercial information that is confidential. Those sections and information/data contained therein which meet these criteria are identified using a "grey" background. Appendices cited which meet these criteria are contained in a sub-folder labelled "Do Not Disclose".

Thomas Berger
Vice President G-Science, Inc.

10/17/2018
Date

TAB

PART 2

PART 2

DETAILED INFORMATION ABOUT THE NOTIFIED SUBSTANCE

Table of Contents

2. DETAILED INFORMATION ABOUT THE NOTIFIED SUBSTANCE

2.1 Identity

Clinoptilolite is a natural zeolite that occurs in sedimentary and volcanic rocks, altered basalts, ores and clay deposits. Zeolites are hydrated aluminosilicates that possess a three-dimensional framework structure. They are characterized by their ability to lose and gain water reversibly, adsorb molecules of appropriate cross-sectional diameter (adsorption or acting as molecular sieves) and exchange their extra-framework cations without major change of their structure (ion exchange property) (Ames, 1960; Sheppard, 1971; Mumpton and Fishman, 1977).

Each zeolite species has its own unique crystal structure, thus its own set of physical and chemical properties. Clinoptilolite is one of the most commonly mined zeolites worldwide (USGS, 2015). It belongs to the heulandite group of tectosilicate minerals and includes three species: clinoptilolite Ca, clinoptilolite Na and clinoptilolite K; with the respective chemical symbol indicating the most dominant extra-framework cation in the chemical formula (Coombs et al., 1997). Factors affecting clinoptilolite cation selectivity include cation size, charge, electronic structure and, in the presence of Na, temperature. Clinoptilolite has a thermal stability of up to 700°C (without reaction at lower temperatures) and a Si:Al ratio of ≥4. The small amount of Al in clinoptilolite results in a relatively low cation exchange capacity (CEC) (~2.3 meq/g), however, clinoptilolite has a decided preference for larger cations, including NH_4^+ (Ames, 1960; Mumpton, 1960), with adsorption capacity increasing with pH (Kithome et al., 1999). The affinity of clinoptilolite for NH_4^+ is the basis for its widespread use in the treatment of animal wastes and municipal/industrial wastewater (Jorgensen et al., 1976; Mumpton, 1985, 1999; Inglezakis, 2005; Jha and Hayashi, 2009). Because of its adsorption, cation exchange, catalysis and dehydration properties, clinoptilolite is used in a variety of applications in the fields of agricultural/animal production, construction, industrial technology, environment and human medicine (Mumpton, 1985; Auerbach et al., 2003; Reháková et al., 2004; Colella, 2011).

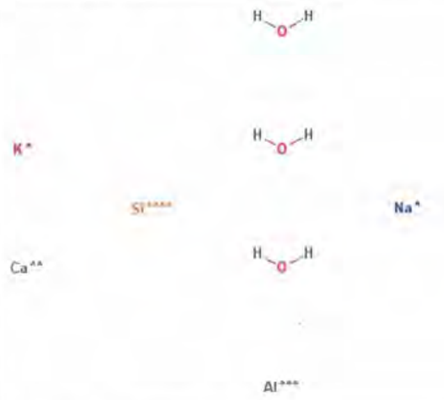
2.2 Characterization of Zeocem Clinoptilolite

The clinoptilolite which is the subject of this GRAS notice is a natural crystalline aluminosilicate mineral of sedimentary origin that is exclusively sourced at Zeocem's Nižný Hrabovec mine in the Prešov Region of the Slovak Republic.

Natural clinoptilolites are characterized primarily by the content of clinoptilolite and secondly by the presence of clays. A commonly accepted mineralogical analysis for clinoptilolite is X-ray diffraction (XRD) (CRL, 2010). Based on this analysis, the Zeocem clinoptilolite has a minimum clinoptilolite content of 80%, a maximum content of 20% accessory minerals and is free of quartz and fibers of other zeolite minerals. Particle size ranges from 0.01 to 2.5 mm (D90[1]). The clinoptilolite is primarily of the species Ca and K, with low content of Fe, Mg and Na ions (Reháková et al., 2004; Domaraká et al., 2015).

The general physical and chemical properties of Zeocem clinoptilolite are summarized Table 2.1. An overview of the composition and the specification is given in Table 2.2. Additional detailed information about its specific qualitative parameters is given in Appendices 2.1, 2.2, 2.6a - 2.10. Detailed information on granulometric data for the Zeocem clinoptilolite described in this notification is described by Reháková et al. (2004). Information on particle size is also given in Section 3.3.1.

[1] The D(x) is the diameter of the particle that x% of a sample's mass is smaller than and (100-x)% of a sample's mass is larger than this value.

Table 2.1	**Physical and chemical properties of clinoptilolite**
Parameter	Description
Clinoptilolite	A natural crystalline aluminosilicate mineral of sedimentary origin containing ≥80% clinoptilolite and ≤20% accessory minerals (free of fibers and quartz)
Chemical formula (as a naturally occurring mineral, the precise composition of clinoptilolite is subject to a degree of variation)	$(Ca, K_2, Na_2, Mg)_4\ Al_8\ Si_{40}\ O_{96}\ 24H_2O$
Molecular weight	211.279 g/mol
Structure	
CAS No.	12173-10-3
Synonyms	Clinoptilolite
Uses	Anticaking agent
Color/Form	Light grayish-green
Odor	None
Physical state @ 20°C	Solid
Solubility in water	None
Melting point	1340°C
Flow temperature	1420°C
Ignition temperature	Settled dust – neg. up to 600 °C, raised dust – neg. up to 800 °C.
Flash point	Neg. up to 600 °C.
Explosive limits (volume % in air)	Non-explosive
Specific gravity	2200-2440 kg/m^3
Density	1600-1800 kg/m^3
Porosity	24 - 32%
Pore diameter	0.4 nm (4 Å)
Comprehensive strength	33 MPa
Mohs hardness	1.5 – 2.5
Grindability index determination	kVTI = 1.628
Specific surface (BET)	30-60 m^2/g
Cation exchange capacity	1.20 – 1.50 mol/kg
Acid stability	79.5%
Thermal stability	up to 400 °C
Source: Appendices 2.1 and 2.2; Vatalová et al (2016); PubChem (2016).	

BEST COPY AVAILABLE

Table 2.2	**Specification of Zeocem clinoptilolite**
Parameter	Content
Clinoptilolite of sedimentary origin	Not less than 80 %
Clay minerals	Not more than 20 %
Particle size	0.01 - 2.5 mm (D90)
Loss on drying	≤ 6%
SiO_2	62 - 73%
Al_2O_3	11 - 14%
Si:Al	4.8 - 5.4
CaO	2 - 5.5%
Na_2O	0.2 - 1.5%
Fe_2O_3	0.7 - 2.3%
K_2O	2.2 - 3.4%
MgO	0.5 - 1.2%
Pb	Not more than 20 mg/kg
Cd	Not more than 0.1 mg/kg
As	Not more than 2 mg/kg
Hg	Not more than 0.05 mg/kg
Dioxins (such as PCDD/F)	Not more than 0.3 ng WHO - TEQ/kg
Dioxins (such as PCDD/F) + Dioxin-like PCBs	Not more than 0.5 ng WHO - TEQ/kg

2.3 Technical Effect

2.3.1 Intended use - anticaking agent

Clinoptilolite is member of the group of zeolites which are crystalline, hydrated aluminosilicates (Sections 2.1 and 2.2). It is an inert material which does not dissolve in water, but may adsorb water (Kotova et al., 2016). Due to its high adsorption rate, cation exchange, catalysis and dehydration capacities, clinoptilolite acts as an anticaking agent that when added to feed keeps it dry and free-flowing. Under normal environmental conditions, clinoptilolite has a stable crystal structure, with mineral-specific ion exchange and adsorption properties and reversible hydration capacity.

2.3.2 Utility/technical effect as anticaking agent

The Code of Federal Regulation includes in section 582 currently three aluminosilicate compounds that are GRAS when used as anticaking agents in animal feed at a use tolerance level of 2% in accordance with good manufacturing or feeding practice. These are Aluminum calcium silicate (21 CFR 582.2122), Sodium aluminosilicate (21 CFR 582.2727), and Hydrated sodium calcium aluminosilicate (21 CFR 582.2729). The same substances are also listed by CFR as being GRAS when used in human food, 21 CFR 182.2122, 182.2727 and 182.2729. There are other silicates such as pyrophyllite or verxite permitted as food additives for use in animal feed as anticaking agents.

The use of GRAS aluminosilicates as anticaking agents in food was reviewed by Peleg and Hollenbach (1984) who also provide some general background on the properties of substances that act as flow conditioners/anticaking agents. Anticaking agents (also known as glidants, anti-agglomerants, lubricants or free-flowing agents) are finely divided solids that are added to a host powder to improve flow and/or reduce its tendency to cake (Hollenbach et al., 1982). They are usually inert materials which do not dissolve in water, but many can adsorb water in large amounts.

The anticaking activity of conditioners is achieved in several ways. One is coating of host powder particles which reduces their clumping, the lubricant separates physically the host particles and reduces friction caused by cohesion. A second important mechanism is competition for adsorbed water where the host powder stays dry and flowable due to the water adsorption capacity of the anticaking agent (Irani et al., 1961). Several other mechanisms may work, for example, as one where the anticaking agent neutralizes electrostatic charges or reduces superficial molecular attractive forces. This mechanism is also important for reducing caking at surfaces of processing equipment. It should be noted that in a given system of a host powder and an anticaking agent these mechanisms may be present jointly and support each other synergistically. This aspect was also emphasized by Adhikari et al. (2001) when discussing stickiness of food, and more recently by Zafar et al. (2017) in their review on bulk powder caking.

For the described physical separation, a particle size between 1 and 50 µm may be desirable (Irani et al., 1959), for fine host powder even very fine conditioner with particle sizes below 0.1 µm may be needed (York, 1975). In case of competition for adsorbed water or other mechanisms particles of larger size will work as well.

Aluminosilicates continue to be suitable anticaking agents for powdered food items such as cake and dessert mixes, egg powder, dairy products, sauce, onion and garlic powders, soup powder, sucrose, corn starch, lactose, modified corn starch (MCS), sucrose and MCS matrix (Peleg and Hollenbach, 1994; Adhikari et al., 2001; Zafar et al., 2017). Many of these powders are also used as feed ingredients or represent matrices that are found in feed ingredients.

The suitability of aluminosilicates used as anticaking agents in food applies therefore to their use in animal feed for the same purpose, that is to be added to finely powdered or crystalline feed ingredients to prevent caking, lumping, or agglomeration.

Zeocem clinoptilolite is a hydrated aluminosilicate with a mineral composition levels of SiO_2 and Al_2O_3 that are similar to levels found in the GRAS aluminosilicates, specifically hydrated sodium calcium aluminosilicate (21 CFR 582.2729).

Clinoptilolite is, like the listed three GRAS aluminosilicates, an inert material which does not dissolve in water, but may adsorb water (Kotova et al., 2016). Clinoptilolite from sedimentary origin is available over a broad range of particle diameters (Reháková et al., 2004) and will support any of the described mechanisms of anti-caking. As a fine powder with diameter in the single digit micron range or even lower it will cover host powder particles, more granular forms with diameters larger than 50 µm will still adsorb water and reduce caking of feeds by keeping them dry.

It can therefore be expected that the Zeocem clinoptilolite when used as an anticaking agent in feed would act similarly to the GRAS anticaking agents currently permitted. Zeocem clinoptilolite meets the EU regulations as an anticaking agent to be added to animal feed for all animal species at a level of 1% (EU, 2013; EC 2018).

The efficacy of an additive as an anticaking agent can be demonstrated by measuring the flowability of feed materials or compound feed, without and with the additive (Jenike, 1964). The European Community (EC) Expert Group for Technical Advice on Organic Production reported that the addition of 2% clinoptilolite to feed compounds improves flow properties (EC, 2011).

2.3.3 Potential interference of clinoptilolite with a medicated feed assay

Clinoptilolites are known to absorb not only water (which is part of the anticaking agent property) but also other components such as heavy metals and ammonia (Minceva et al., 2008; Margeta et al., 2013; Taparcevska et al., 2010). It is known from other clays, e.g. bentonite, that they may interfere with medication (EFSA, 2012b). Bentonite is used or intended for use animal feed as an anti-caking agent and pelleting aid in an amount not to exceed 2% in the total ration (AAFCO, 2016). It is considered GRAS (21 CFR 582.1155) and can be used in food with no limitation other than current good manufacturing practice. Bentonite is not prohibited in medicated animal feed for the same purposes and at the same levels when it can be demonstrated that it does not interfere with the bioavailability of the medicament to animals and the analysis of the feed for the medicament by acceptable methods. Medicaments with which bentonite can and cannot be currently used are listed in Section 73.1 of AAFCO (2016).

There are no studies that address potential interference of feeding clinoptilolite (as an anticaking agent or otherwise) with the periodic assay of medicated feed. It is useful to note that even in the EFSA (2013) scientific opinion report on the safety and efficacy of clinoptilolite of sedimentary origin for all animal species, it is stated that a structured database search to provide information on target animal safety and interactions with other feed components was carried out. There is no mention in the report of any studies involving the feeding of clinoptilolite and drug interference.

Several studies were found that describe the use of clinoptilolite as a drug carrier where its interaction with medicaments or organic molecules of pharmaceutical interest was investigated using experimental and theoretical tools. These studies are highlighted below.

Lam et al. (1998) carried out theoretical study on the physical absorption of aspirin on natural clinoptilolite. They reported that the aspirin molecule will be adsorbed in a stronger fashion in an acid zeolite than in a sodium zeolite. However, they were not able to find any experimental results or theoretical calculations with which to compare their findings. Lam et al. (2001) conducted a similar study with natural clinoptilolite and metronidazole. The results obtained suggested a possible adsorption of metronidazole on clinoptilolite.

Farias et al. (2003) carried out interaction studies between a natural clinoptilolite and some of its modified forms with two drugs (metronidazole and sulfamethoxazole) within a wide range of pH values of pharmaceutical interest (1.2, 3.0, 5.5 and 8) by using UV and IR spectroscopy. It was demonstrated that the organic molecules do not show signals of degradation after the contact with the different clinoptilolite products at any pH values. Likewise, the interaction between clinoptilolite and sulfamethoxazole was found to be negligible at the pH values considered.

The use of clinoptilolite as a drug carrier for other antibiotics has also been investigated for erythromycin (Cerri, 2004) and cephalexin (Nezamzadeh-Ejhieh and Tavakoli-Ghinani, 2014).

Despite the lack of scientific information concerning potential interference of clinoptilolite with an assay of medicated feed, the Notifier understands that it is the feed manufacturer's responsibility to determine and provide adequate data to support the conclusion that such interference does not occur before using the product in a specific feed. In this regard, it is proposed that the approach currently in effect for bentonite (IFN-8-00-695), a naturally occurring mineral consisting primarily of the tri-layered aluminum silicate (montmorillonite) may be applied also for clinoptilolite: when using it in medicated feed, it may be necessary to demonstrate that it does not interfere with the bioavailability of the medicament to animals and the analysis of the feed for the medicament by acceptable methods. A corresponding statement would be added to the product label for clinoptilolite.

2.4 Stability Data

The clinoptilolite is certified by Zeocem as being stable for 24 months from the manufacturing date when stored in original, undamaged and closed containers in dry, hygienically clean, well-aired indoor storage facilities. The product is packaged in paper bags (25 kg) shipped as 40 bags/pallet and "BigBag" packaging (e.g. 1,000 kg/1,200 kg/pallet) or shipped as bulk material for storage in silos (Appendix 2.1). Stability data for one a production batch of Zeocem clinoptilolite over a four-year period is given in Table 2.4.

Table 2.4 Stability of Zeocem clinoptilolite over four-year period (Batch M 070103-1)

Product	Sample no.	Testing date	Loss on drying[a] (%)	Exchange capacity[b] (mol/kg)	Cumulative oversize[c] (%) 0.09 mm	Cumulative oversize[c] (%) 0.2 mm	Months of storage
Zeocem clinoptilolite (25 kg bags)	1	08.01.2003	3.52	(b) (4) [redacted]			
	2	08.07.2003	3.55				
	3	09.01.2004	3.54				
	4	09.07.2004	3.55				
	5	10.01.2005	3.54				
	6	08.07.2005	3.55				
	7	09.01.2006	3.55				
	8	10.07.2006	3.56				
	9	08.01.2007	3.57				

Standards used:
[a] STN 720102 - Basic analysis of silicates. Determination of loss by drying.
[b] CSN 721076 - Determination of exchange capacity and exchangeable cations of clay soils. (Note: the applied method determines partial exchange capacity – in Table 2.1. the Cation exchange capacity is higher as it is determined differently)
[c] STN 721213 - Physical and mechanical tests of limestone (sieving analysis-wet way).

2.5 Homogeneity Data

The efficacy of clinoptilolite as an anticaking agent was discussed in terms of flowability of feed materials (Section 2.3.2). The free flow of feed can only be achieved if an anticaking agent is distributed homogenously (otherwise clumps would occur); therefore, homogeneity studies are not necessary.

2.6 Manufacturing Process for the Notified Substance

2.6.1 Mined and processed raw material

The Zeocem clinoptilolite is exclusively sourced at the Nižný Hrabovec mine in the Prešov Region of the Slovak Republic. The raw material is excavated using conventional pit mining techniques. Drill core samples of the exposed ore are taken prior to blasting to determine action exchange capacity (CEC). After blasting of the selected area, the raw material is transported to an on-site Jaw crusher where the material is crushed to a size ranging from 25 to 160 mm. Any fractions between 0 to 25 mm are removed by means of a coarse screen. The sorted materials are then stored in steel silos (300 m3 capacity) until being transported in bulk by trucks to the manufacturing site in Bystré and deposited in a covered storage area. Some homogenization of the material takes place during the loading, transport and dumping stages. The stored bulk material is rechecked daily for CEC and loss on drying. Any material failing to meet the standard requirements is then removed following a plan of set methods for control of incongruous material. The accepted material is then dried at 160 °C in the two modern gas dryers until the loss on drying is ≤ 6% and then stored in bulk in a covered area. Loss on drying is checked twice per shift (three shifts per day).

A flow diagram depicting the different steps in the manufacturing of Zeocem's ground and granular clinoptilolite products is given in Appendix 2.4. The production process for the granular form of clinoptilolite is described below.

(b) (4)
[redacted]

(b) (4) [redacted]

2.6.2 Traceability of product

Traceability of each production batch is carried out according to recognized industry practices. Each production batch is coded as per one of three formats (Table 2.5).

Table 2.5. Production batch codes used for products

Code	Definitions of code
(b) (4) [redacted]	(b) (4) [redacted]

2.6.3 Process controls

(b) (4) [redacted]

(b) (4)

[redacted]

2.6.4 Certificates of registration

Code of practice and good feed manufacturing practices certificates issued to Zeocem by the feed industry's quality/safety system for specialty feed ingredients and their mixtures (FAMI-QS) and the Central Controlling and Testing Institute in Agriculture in Bratislava are given in Appendices 2.11 and 2.12. A material safety data sheet (MSDS) for clinoptilolite is given in Appendix 2.13 and an example of a Zeocem ZeoFeed® product label in Appendix 2.14.

TAB

PART 3

PART 3

TARGET ANIMAL AND HUMAN EXPOSURE

Table of Contents

3. TARGET ANIMAL AND HUMAN EXPOSURE

3.1 Target Animals

3.1.1 Intended levels of use

Zeocem clinoptilolite is intended to be added to a complete feed mixture or to feed premixes of the diets of livestock (ruminants and swine), poultry and pets (dogs and cats) at a level not exceeding 1% (10,000 mg/kg feed) by weight of the feed composition in accordance with good manufacturing or feeding practice.

3.1.2 Mineral substances and impurities

Clinoptilolite is found in sedimentary and volcanic rocks that formed under diverse geological occurrence. Therefore, it also contains minor amounts of other minerals and impurities. These substances are therefore also consumed by the animal when clinoptilolite is added to the diet. An overview of the qualitative parameters of Zeocem clinoptilolite (oxide minerals, heavy metals, dioxins, radioactive and microbial contamination, etc) is given in Appendices 2.1, 2.2 and 2.10. Examples of detailed analyses carried out are given for the content of dioxins (Appendix 3.1a), radionuclides (Appendix 3.3) and constituents of animal origin (Appendix 3.4).

3.1.3 Oxides, arsenic and heavy metals

The specifications of Zeocem clinoptilolite and percentage range in values of its compositional content were given in Table 2.2. The daily amounts of clinoptilolite and additional oxide mineral substances and heavy metals that would be consumed by target animals when the anticaking agent is added to the diet at the maximum recommended level of 1% are given in Table 3.2. These amounts are then compared to the respective maximum tolerable levels of these substances as recommended by NRC (2005) and AAFCO (2016) (Table 3.3). The data demonstrate that the amounts of mineral substances and heavy metals that target animals would ingest when consuming diets containing 1% clinoptilolite are well below (e.g. >100 times less for Pb and Hg) the recommended tolerable levels. The element with the highest level in the diet is aluminum, but the amount consumed would be nearly 25% less than the maximum level of 1000 mg/kg proposed by (NRC, 2005). With respect to aluminum it should be noted that the NRC guidance level is derived from studies with soluble aluminum salts where aluminum is bioavailable; as discussed in Part 6.4.1 aluminum present in clinoptilolite is de facto not bioavailable (aluminum is present in clinoptilolite as an insoluble oxide and not as a soluble salt).

3.1.4 Dioxins and dioxin-like PCBs

US regulatory levels for dioxins and dioxin-like polychlorinated biphenyls (PCBs) in food or feed have not been officially established (Institute of Medicine, 2003). However, provisional levels of PCBs of 0.2 ppm are permitted by the FDA in finished animal feed for food-producing animals, except feed concentrates, feed supplements and feed premixes; and 2 ppm in animal feed components of animal origin, including fishmeal and other by-products of marine origin and in finished animal feed concentrates, supplements, and premixes intended for food-producing animals (21 CFR 509.30).

In the EU tolerances of 0.75 ng WHO-TEQ/kg (ppt) for dioxins and 1.5 ng WHO-TEQ/kg (ppt) for the sum of dioxin and dioxin-like PCBs are enforced for complete feed and clinoptilolite (EC, 2013).

Zeocem clinoptilolite contains dioxins and dioxin-like PCBs at levels typically below 0.3 ppt, the average content is 0.15 ppt (Appendices 3.1 and 3.2). At the proposed use level, the contribution to complete feed is 0.003 ppt. This is at least one order of magnitude lower than the lower side of the range of reported levels of contamination in feed ingredients summarized by the Institute of Medicine (US) Committee on the Implications of Dioxin in the Food Supply (Institute of Medicine, 2003).

The intake of dioxins and dioxin-like PCBs by target animals consuming feed containing Zeocem clinoptilolite is very low as demonstrated by the daily intake estimates for target animals given in Table 3.3.

3.1.5 Radionuclide activity

Zeocem clinoptilolite is regularly monitored for presence of ^{137}Cs (0.6 - 2.3 Bq/kg) and ^{134}Cs (not detected). Batch data on ^{40}K (1179.20 Bq/kg), ^{226}Ra (45.6 Bq/kg), and ^{232}Th (56.4 Bq/kg) is given in Appendix 3.3.

The levels for caesium are well below the FDA guidance levels for food (CPG 7119.14). The levels for the three natural radionuclides will, at the use level of 1% clinoptilolite, contribute little to total intake as these radionuclides are known to be present in other feed ingredients (Wiechen 1998; ATDSR, 1990a,b). Intake estimates of radionuclides by target animals are given in Table 3.4.

3.1.6 Comparison of contaminants in Zeocem clinoptilolite vs other clinoptilolites

The clinoptilolite subject to the present GRAS notice may differ not only in the major mineral constituents, but also the contaminants (heavy metals, dioxins and PCBs) from other clinoptilolites used in the studies reviewed in Part 6 of the original Notice. Such comparison is not easy as the inclusion of such detailed analytical data for clinoptilolites used in feeding trials published in peer-reviewed journals is not routine. Often only the purity of the clinoptilolite and percentage that is fed is given, and in some cases, the percentages of SiO_2, Al_2O_3 and other oxides may be provided. Therefore, because of the lack of such information, it is not possible to directly compare the contaminants in all clinoptilolites used in the studies reviewed to Zeocem clinoptilolite. The following considerations are based, in the absence of US tolerances, on the maximum limits of the EU which reflect *inter alia* international evaluations by JECFA.

Maximum limits for heavy metals, arsenic, dioxins and dioxin-like PCBs were applied historically in Europe (many as early as 1974, most of them before 2003) and apply today for complete feed, complimentary feed (mineral feed), and for clinoptilolite (EC, 1999, 2002; EU, 2013). These levels are regularly reviewed and continue to reflect state-of-the-art toxicology and good manufacturing practice, including reported and observed ranges of contamination. A comparison of the EU allowable limits of contaminants with levels recommended by NRC (2005) is given in Table 3.1.

The Notifier would expect that levels of contaminants in the complete feeds used in the studies reviewed in Section 6.2 of this Notice are meeting the legal requirements laid down in European Union Feed law. This applies specifically to the studies run in Europe reported by Katsoulos et al (2005a, 2005b, 2005c), Karatzia et al (2011), Pourliotis et al (2012), Papaioannou et al (2002), Sardi et al (2002) and Herzig et al (2008). The purpose for carrying out such studies is to develop data so that a product can be put into the marketplace. Therefore, it stands to reason that the product complies with established regulations so that it can be marketed and sold. Reviewed clinoptilolite studies conducted outside of the EU (including the USA) under a different feed law jurisdiction would, nevertheless, also support the conclusions on the safety of Zeocem clinoptilolite. If the results obtained from such studies do not indicate any safety issues when the respective clinoptilolites were fed at a use level of 1% or higher, then it can be assumed with relative confidence that the study material was not contaminated at levels which would result in toxicity.

Table 3.1	**Tolerance levels for selected feed contaminants in the EU and USA**					
	Lead	Cadmium	Mercury	Arsenic	Dioxins	Dioxin-like PCBs
EU [a]	mg/kg				ng WHO - TEQ/kg	
Complete feed	5	1	0.1	2	0.75	1.5
Complementary feed	10	0.5	0.2	4	0.75	1.5
Mineral feed	15	5	0.2	12	NA	NA
Clinoptilolite	30	2	[0.1]	[2]	0.75	1.5
USA [b]	10 – 100	10	0.2	30	NA	NA

[a] EC (1999, 2002).

[b] NRC (2005).
[ ] Levels not legally mandatory, but respected and applied by the feed chain.
NA - Not available.

In Part 6.2 of this Notice data from target animal studies where clinoptilolite from other mines had been used is presented. As the purity of the Zeocem clinoptilolite is ≥80%, emphasis has been given to reviewing studies in which the purity of clinoptilolite is stated as being at least 80%. In Part 6.3.4 the similarities between the composition of Zeocem clinoptilolite and the composition of the different clinoptilolites used in six feeding studies is summarized (Table 6.1). The data presented clearly demonstrate that the purity and mineral composition of the oxides of the Zeocem clinoptilolite is very similar to that of the clinoptilolites used in the reported European studies, and for which no safety concerns were indicated for toxicity due to contaminants. Likewise, the similarities in the mineral composition of Zeocem clinoptilolite and the composition of the clinoptilolites used in two safety trials (one conducted in the USA) are shown in Table 6.3.

Therefore, the Notifier believes that the clinoptilolites used in European studies are similar to the Zeocem clinoptilolite with respect to their likely contamination with heavy metals, arsenic and dioxins/dioxin-like PCBs, as they all were products used under applicable EU feed law which stipulates levels for these contaminants (EFSA, 2012a; EU, 2013). The safety data from animal feeding studies reviewed in Part 6.2 demonstrates that clinoptilolite can be added to the diets of ruminants, swine and poultry under normal production conditions at levels ranging from 1 to 7.5% without eliciting detrimental effects on animal health or overall performance. The Notifier is not aware of any animal feeding studies with clinoptilolite where toxicity was reported due to presence of contaminants, and therefore considers that the studies cited in Part 6.2 are sufficiently similar with respect to their spectrum and level of contaminants to Zeocem clinoptilolite.

3.2 Human Exposure

As is described in detail in Part 6 (Section 6.4.1), there is no evidence that clinoptilolite is degraded during its passage through the gastrointestinal tract of target animals. The physiological and chemical conditions in the animal's digestive system (pH, digestive enzymes, etc) are not enough to decompose clinoptilolite; the substance is essentially not absorbed and is excreted with the faeces. No scientific reports were found demonstrating that any impurities (minerals or heavy metals) contained in clinoptilolite complying with the specifications as laid down in Part 2 of this Notice were deposited in tissues of animals to which the substance was fed in amounts above maximum permitted levels. Even clinoptilolite with a high level of lead (46 mg/kg), when added at a level of 2% to pigs feed for 45 days, resulted in a low retention coefficient of lead from clinoptilolite of 0.009 (Fokas et al, 2004).

Therefore, based on the information available at the time of this submission, it can be stated that human exposure to residues of clinoptilolite in animal tissues is unlikely to pose a potential risk or safety issue (Section 6.5).

Table 3.2		**Target animal exposure to clinoptilolite (upper limits)**									
Animal category [a]		Daily feed intake [b] (kg)	Daily clinoptilolite Intake [c] (g)	Daily intake of minerals, arsenic and heavy metals contained in clinoptilolite [d]							
				Silicon (g)	Aluminum (g)	Iron (g)	Calcium (g)	Lead (mg)	Arsenic (mg)	Cadmium (µg)	Mercury (µg)
Ruminants	Calves (≤15 months, 300 kg)	7.10	71.00	24.23	5.26	1.14	2.79	1.42	0.142	7.100	3.550
	Dairy cows (≥2 years, 680 kg)	26.90	269.00	91.79	19.93	4.33	10.57	5.38	0.538	26.900	13.450
	Sheep (≥8 months,140 kg)	1.98	19.80	6.76	1.47	0.32	0.78	0.40	0.040	1.980	0.990
	Goats (≥8 months, 90 kg)	1.54	15.40	5.25	1.14	0.25	0.61	0.31	0.031	1.540	0.770
Swine	Piglets (3.5 - 5 kg)	0.25	2.50	0.85	0.19	0.04	0.10	0.05	0.005	0.250	0.125
	Piglets (5.0 - 10 kg)	0.50	5.00	1.71	0.37	0.08	0.20	0.10	0.010	0.500	0.250
	Piglets (10 - 20 kg)	1.00	1.61	0.55	0.12	0.03	0.06	0.03	0.003	0.161	0.080
	Pigs (20 - 50 kg)	1.86	18.55	6.33	1.37	0.30	0.73	0.37	0.037	1.855	0.928
	Pigs (50 - 80 kg)	2.58	25.75	8.79	1.91	0.41	1.01	0.52	0.052	2.575	1.288
	Pigs (80 - 120 kg)	3.08	30.75	10.49	2.28	0.49	1.21	0.62	0.062	3.075	1.538
	Sows (lactating) (175 kg)	5.25	52.50	17.91	3.89	0.84	2.06	1.05	0.105	5.250	2.625
Poultry	Layers (commercial/breeder)	0.10	1.00	0.34	0.07	0.02	0.04	0.02	0.002	0.100	0.050
	Broilers (starter, 0-3 weeks)	0.07	0.66	0.23	0.05	0.01	0.03	0.01	0.001	0.066	0.033
	Broilers (grower, 4-6 weeks)	0.15	1.53	0.52	0.11	0.02	0.06	0.03	0.003	0.153	0.077
	Broilers (finisher, 7-9 weeks)	0.20	2.02	0.69	0.15	0.03	0.08	0.04	0.004	0.202	0.101
	Turkeys (starter, 0-8 wk)	0.21	2.10	0.72	0.16	0.03	0.08	0.04	0.004	0.210	0.105
	Turkeys (grower, 8-16 wk)	0.49	4.90	1.67	0.36	0.08	0.19	0.10	0.010	0.490	0.245
	Turkeys (finisher, 16-24 wk)	0.75	7.50	2.56	0.56	0.12	0.29	0.15	0.015	0.750	0.375
Pets	Dogs (adult 15 kg)	0.21	2.10	0.72	0.16	0.03	0.08	0.04	0.004	0.210	0.105
	Cats (adult 3 kg)	0.04	0.42	0.14	0.03	0.01	0.02	0.01	0.001	0.042	0.021

[a,b] Animal categories (age, weight and/or production) and respective daily feed intake are based on NRC (1994, 1998, 2000, 2001, 2006, 2007) and FEDIAF (2013).
[c] Use level of 1% of diet by weight.
[d] Calculated based on data in Table 2.2 and Appendices 2.1, 2.2 and 2.6a - 2.10.

Table 3.3 **Levels of minerals and heavy metals in diets supplemented with Zeocem clinoptilolite compared to recommended tolerable levels**[a]
Silica. Insoluble forms of silica can be tolerated by livestock at levels as high as 50,000 mg/kg diet (NRC, 2005). Silica dioxide is considered to be GRAS (21 CFR 573.940) and (21 CFR 582.80). Assuming a maximum level of 73% SiO_2, adding clinoptilolite to diets at a level of 1% would result in silica being present at a level of 3412 mg/kg diet, or nearly 15 times less than the maximum tolerable level.
Aluminum. It is estimated that more soluble forms of aluminum can be tolerated at levels of 1000 mg/kg diet (NRC, 2005; AAFCO, 2016). Assuming a maximum level of 14% Al_2O_3, adding clinoptilolite to diets at a level of 1% would result in aluminum being present at a level of 741 mg/kg diet which is below the maximum tolerable level.
Iron. The proposed maximum safe level of iron is 500 mg/kg for ruminants and poultry, and 3000 mg/kg for swine (NRC, 2005). EFSA FEEDAP Panel recommended a maximum safe content of iron in complete feed as being 450 mg/kg for cattle, 500 mg/kg for ovines, 600 mg/kg for cats, dogs and poultry, and 3000 mg/kg for pigs (EFSA, 2016). Iron oxide is listed to be GRAS when added to food (21 CFR 186.1374) and feed (21 CFR 582.80) as a nutritional dietary supplement at levels consistent with good feeding practice. Assuming a maximum level of 2.3% Fe_2O_3, adding clinoptilolite to diets at a level of 1% would result in iron being present at a level of 161 mg/kg diet. This amount is well within the recommended ranges given by NRC (2005) and EFSA (2016).
Calcium. NRC (2005) recommends upper tolerable levels of calcium for ruminants and poultry as being 1.5% of daily diet, for swine 1.0% of daily diet, 2% of daily intake for dogs and 1% for cats. As an example, for an adult dairy cow this would be equivalent to about 404 g/d of Ca or 15000 mg/kg of diet. Calcium oxide is listed to be GRAS when added to food (21 CRF 184.1210) and feed (21 CFR 582.1210) as a nutritional dietary supplement at levels consistent with good feeding practice. Assuming a maximum level of 5.5% CaO, adding clinoptilolite to diets at a level of 1% would result in calcium being present at a level of 393 mg/kg diet, or about 38 times less than the maximum tolerable level.
Lead. Studies have demonstrated that cattle, sheep and poultry can tolerate 10 mg/kg diet for extended periods without exhibiting adverse effects, however, actual maximum tolerable levels have not been established for livestock species (NRC, 1980, 2005). It is recommended that of levels of lead in animal diets be limited to 30 mg/kg diet (NRC, 1980; AAFCO, 2016). Assuming a maximum level of 20 mg/kg Pb, adding clinoptilolite to diets at a level of 1% would result in lead being present at a level of 0.2 mg/kg diet, or about 150 times less than the maximum recommended level of 30 mg/kg diet.
Cadmium. Dietary levels of cadmium (10 mg/kg) are tolerated chronically by poultry and ruminant species, but these levels result in unacceptable levels of cadmium in tissue and muscle. The World Health Organization has set a 1 mg/kg upper limit for cadmium in complete feeds for animals (NRC, 2005). AAFCO (2016) proposes 0.5 mg/kg diet. If present at the maximum specified level of 0.1 mg/kg Cd, adding clinoptilolite to diets at a level of 1% would result in incremental cadmium being added to feed at a level of 0.001 mg/kg diet, or 500 times less than the accepted upper limits.
Arsenic. The suggested maximum tolerable levels of arsenic for domestic livestock proposed by NRC (2005) and AAFCO (2016) are 30 mg/kg and 50 mg/kg diet, respectively. Assuming a maximum level of 2 mg/kg As, adding clinoptilolite to diets at a level of 1% would result in incremental arsenic being added to feed at a level of 0.02 mg/kg diet, or 1500 times less than the accepted upper limits.
Mercury. NRC (2005) and AAFCO (2016) recommend a maximum mercury level of 2 mg/kg diet for ruminants, poultry and pigs. Assuming a maximum level of 0.05 mg/kg Hg, adding clinoptilolite to diets at a level of 1% would result in incremental mercury being added to feed at a level of 0.005 mg/kg diet, or 400 times less than the accepted upper limits.
[a] Amounts of minerals and heavy metals (mg/kg feed) were calculated using the upper ranges of the specification data for clinoptilolite given in Table 2.2 and feed intake data based on NRC (1994, 1998, 2000, 2001, 2006, 2007) and FEDIAF (2013). The levels of minerals and heavy metals presented in Tables 3.2 only reflect those amounts that would be contained in the diet due to the addition of 1% clinoptilolite and do not include any potential impurities present in feed prior to adding the anticaking agent.

Table 3.4 Target animal exposure to clinoptilolite (upper limits)

Animal category [a]		Daily feed intake [b] (kg)	Daily clinoptilolite intake [c] (g)	Amounts of dioxins and radionuclides contained in respective amounts of clinoptilolite consumed per day [d]									
				Sum of dioxin and dioxin-like PCBs		Potassium (40)		Radon (226)		Thorium (232)		Caesium (137)	
				ng/d	ng/kg feed	Bq/d	Bq/kg feed	Bq/d	Bq/kg feed	Bq/d	Bq/kg feed	Bq/d	Bq/kg feed
Ruminants	Calves (≤15 months, 300 kg)	7.10	71.00	0.01065	0.002	84	12	3	0	4	1	0.043	0.01
	Dairy cows (≥2 years, 680 kg)	26.90	269.00	0.04035	0.002	317	12	12	0	15	1	0.161	0.01
	Sheep (≥8 months,140 kg)	1.98	19.80	0.00297	0.002	23	12	1	0	1	1	0.012	0.01
	Goats (≥8 months, 90 kg)	1.54	15.40	0.00231	0.002	18	12	1	0	1	1	0.009	0.01
Swine	Piglets (3.5 - 5 kg)	0.25	2.50	0.00038	0.002	3	12	0	0	0	1	0.002	0.01
	Piglets (5.0 - 10 kg)	0.50	5.00	0.00075	0.002	6	12	0	0	0	1	0.003	0.01
	Piglets (10.0 - 20.0 kg)	0.22	2.20	0.00033	0.002	3	12	0	0	0	1	0.001	0.01
	Pigs (20 - 50 kg)	1.86	18.55	0.00278	0.002	22	12	1	0	1	1	0.011	0.01
	Pigs (50 - 80 kg)	2.58	25.75	0.00386	0.002	30	12	1	0	1	1	0.015	0.01
	Pigs (80 - 120 kg)	3.08	30.75	0.00461	0.002	36	12	1	0	2	1	0.018	0.01
	Sows (lactating) (175 kg)	5.25	52.50	0.00788	0.002	62	12	2	0	3	1	0.032	0.01
Poultry	Layers (commercial/breeder)	0.10	1.00	0.00015	0.002	1	12	0	0	0	1	0.001	0.01
	Broilers (starter, 0-3 weeks)	0.07	0.66	0.00010	0.002	1	12	0	0	0	1	0.000	0.01
	Broilers (grower, 4-6 weeks)	0.15	1.53	0.00023	0.002	2	12	0	0	0	1	0.001	0.01
	Broilers (finisher, 7-9 weeks)	0.20	2.02	0.00030	0.002	2	12	0	0	0	1	0.001	0.01
	Turkeys (starter, 0-8 wk)	0.21	2.10	0.00032	0.002	2	12	0	0	0	1	0.001	0.01
	Turkeys (grower, 8-16 wk)	0.49	4.90	0.00074	0.002	6	12	0	0	0	1	0.003	0.01
	Turkeys (finisher, 16-24 wk)	0.75	7.50	0.00113	0.002	9	12	0	0	0	1	0.005	0.01
Pets	Dogs (adult 15 kg)	0.21	2.10	0.00031	0.002	2	12	0	0	0	1	0.001	0.01
	Cats (adult 3.0 kg)	0.04	0.42	0.00006	0.002	0	12	0	0	0	1	0.000	0.01

[a,b] Animal categories (age, weight and/or production) and respective daily feed intake are based on NRC (1994, 1998, 2000, 2001, 2006, 2007) and FEDIAF (2013).

[c] Use level of 1% of diet by weight.

[d] For dioxins the average content of 0.15 ppt was used, for the radionuclides: ^{40}K: 1179.2 Bq/kg ^{226}Ra: 45.6 Bq/kg ^{232}Th: 56.4 Bq/kg ^{137}Cs: 0.6 Bq/kg.

TAB

PART 4

PART 4

SELF-LIMITING LEVELS OF USE

Table of Contents

4. SELF-LIMITING LEVELS OF USE

As stated in Section 1.4, the levels at which Zeocem clinoptilolite should be added to the diets of livestock (ruminants, swine), poultry, and pets (dogs and cats) should not exceed the recommended guidance level of 1% (10,000 mg/kg feed) by weight of the feed composition in accordance with good manufacturing or feeding practice.

Data reported in studies reviewed in Part 6 (Narrative) demonstrate that clinoptilolite can be added to the diets of livestock and poultry at levels ranging from 3% to 8% of the diet by weight without negatively affecting intake. No studies could be found where these species were fed higher levels of clinoptilolite.

In toxicological studies carried out with mice it was reported that feeding clinoptilolite at levels of 25% to normal mice and 25% and 50% to tumor-bearing animals did not illicit a negative effect on feed consumption. Clinoptilolite ingestion was well tolerated as judged by comparable body masses of control and experimental animals (Martin-Kleiner et al., 2001). Similarly, in a study with rats fed diets containing 25% clinoptilolite no adverse effects on feed intake was reported (Pavelić et al., 2001).

There is no established self-limiting level of use. At what level of the diet clinoptilolite would have to be added to make it self-limiting (unpalatable or technologically impractical) is not known.

TAB

Part 5

Part 5

EXPERIENCE BASED ON COMMON USE IN FOOD BEFORE 1958

Table of Contents

5. EXPERIENCE BASED ON COMMON USE IN FOOD BEFORE 1958

This section is not applicable because the GRAS status of clinoptilolite is based on scientific procedures in accordance with the Code of Federal Regulations (CFR) 21 CFR 570.30(a) and (b).as stated in Part 1, Section 1.5.

TAB

Part 6

Part 6

DETAILED SUMMARY OF THE BASIS FOR THE NOTIFIER'S GRAS DETERMINATION

Table of Contents

6. NARRATIVE

This narrative provides a basis for the Notifier's conclusion that the notified substance clinoptilolite, a natural crystalline aluminosilicate mineral of sedimentary origin, mined by Zeocem, is GRAS when used as an anticaking agent added to complete feed mixtures or feed premixes of diets of livestock (ruminants, swine), poultry and pets (dogs and cats), regardless of stage of production, at a level not exceeding 1% (10,000 mg/kg feed) by weight of the feed composition in accordance with good manufacturing or feeding practice.

The scientific data and information presented in the following sections demonstrate that clinoptilolite is safe under the conditions of intended use for both the target animals and for humans consuming human food derived from food-producing animals consuming the substance. The data and information used to establish this conclusion are referenced within the narrative using an author/date format or appendix number. Full citations for all references are given in Part 7 of this GRAS notice; those references that are generally available are listed in Section 1 and those that are not generally available in Section 2.

We have reviewed the scientific data and information that is available and are not aware of any data and information that are, or may appear to be, inconsistent with our conclusion that the substance clinoptilolite merits GRAS status.

There is no information or data contained in this narrative which is considered by the Notifier to be exempt from the Freedom of Information Act. Likewise, no non-public, safety-related data or information was used in reaching the conclusion of GRAS status for clinoptilolite.

6.1 Clinoptilolite and chemically-related compounds with GRAS status

As described in Part 2, clinoptilolite is a natural crystalline aluminosilicate substance belonging to the class of minerals known as silicates. The chemical composition and purity of the clinoptilolite manufactured by Zeocem as an anticaking agent is well defined (Appendices 2.1 and 2.2).

In this section information provided by FDA and other recognized food/feed regulatory bodies/organizations concerning the composition and purity of compounds listed as being GRAS when used as anticaking agents are reviewed; aluminosilicate compounds are emphasized.

6.1.1 Food and Drug Administration

The 1979 report of the FDA Select Committee of GRAS Substances (SCOGS) reevaluated the safety of six silicate compounds authorized for use as anticaking agents in food (LSRO, 1979; FDA, 2015). These compounds had received GRAS status "through experience based on their common use in food" following the passage of the 1958 Food Additives Amendment to the Federal Food, Drug and Cosmetic Act (GPO, 1958). Three of these compounds are aluminosilicates: aluminum calcium silicate (21 CFR 182.2122), sodium aluminosilicate (21 CFR 182.2727) and sodium calcium aluminosilicate, hydrated (21 CFR 182.2729). The SCOGS report gives a description for sodium aluminosilicate as shown below. No information is provided for the other two aluminosilicate compounds.

Sodium aluminosilicate:
Composition - SiO_2 (≥66%, but ≤71% after drying), Al_2O_3 (≥9%, but ≤13% after drying), Na_2O (≥4%, but ≤7% after drying). Loss on drying ≤8%. Loss after ignition ≥8%, but ≤11%.
Limits of impurities - arsenic ≤3 ppm and heavy metals as lead ≤10 ppm.

The FDA has designated a use tolerance level of 2% in accordance with good manufacturing practice for all three of these anticaking agents when used in food, but no description of the actual composition or purity is given (FDA, 2016a).

The FDA considers these same three aluminosilicate compounds to be GRAS when used as anticaking agents in animal feed at a use tolerance level of 2% in accordance with good manufacturing practice

(21 CFR 582.2122, 21 CFR 582.2727 and 21 CFR 582.2729, respectively) (FDA, 2016b). However, definitions of substance composition or purity for these compounds are lacking as shown by the example of 21 CFR 582.2729 given below (GPO, 2016b).

a) Product. Hydrated sodium calcium aluminosilicate (sodium calcium silicoaluminate). b) Tolerance. This substance is generally recognized as safe for use at a level not exceeding 2% in accordance with good manufacturing or feeding practice.

6.1.2 Food Chemicals Codex

The Food Chemicals Codex (FCC) compendium (developed by the National Academy of Sciences and first published in 1966) includes sodium aluminosilicate (CAS No. 1344-00-9, INS 554) within its listing of chemical description monographs. It gives a general definition of composition as shown below. No information is given for aluminum calcium silicate or hydrated sodium calcium aluminosilicate (FCC, 2010).

Sodium aluminosilicate: Composition - occurs as a fine powder, or as beads. It is a series of hydrated sodium aluminum silicates having Na_2O: Al_2O_3: SiO_2 molar ratios of approximately 1:1:13, respectively. Solubility - insoluble in water and in alcohol and other organic solvents, but at 80°C to 100°C, it is partially soluble in strong acids and solutions of hydroxides.

6.1.3 Association of American Feed Control Officials

The Official Publication of the Association of American Feed Control Officials (AAFCO) contains the most complete listing of feed ingredients and their definitions commonly used in the animal food manufacturing industry (AAFCO, 2016). Under FDA's Center for Veterinary Medicine (CVM) Compliance Policy Guide CPG 665.100 (Common or Usual Names for Animal Feed Ingredients), the definitions, as they appear in the Official Publication, are generally regarded as constituting the common or usual name for animal food ingredients, including pet food (FDA, 1995). AAFCO lists the same three aluminosilicates used as anticaking agents in feed as the FDA (21 CFR 582.2122, 21 CFR 582.2727 and 21 CFR 582.2729, respectively). Again, no information is given regarding their composition or purity (AAFCO, 2016).

6.1.4 FAO/WHO

The Food and Agriculture Organization (FAO) and World Health Organization (WHO) joint international body for food standards - Codex Alimentarius (Food Code) and the Joint FAO/WHO Expert Committee on Food Additives (JECFA) list sodium aluminosilicate (INS 554) and calcium aluminium silicate (INS 556) as anticaking agents in food (WHO, 1984; FAO, 2005a,b; CODEX, 2016). The criteria for composition and purity for these two compounds are given as follows:

Sodium aluminosilicate: Composition - a series of hydrated sodium aluminium silicates. The article of commerce may be specified further to silicon dioxide, aluminium oxide, and sodium oxide content, loss on drying, loss on ignition and pH slurry in water. Limits of impurities - lead (≤5 mg/kg). Solubility - insoluble in water and ethanol, partially soluble in strong acids and alkali hydroxides.
Aluminium calcium silicate: Composition - SiO_2 (≥44%, but ≤50%), Al_2O_3 (≥3%, but ≤5%), CaO (≥32%, but ≤38%), Na_2O (≥ 0.5%, but ≤4%). Loss on ignition ≥14%, but ≤18%. Loss on drying ≤10%. Limits of impurities - fluoride ≤50 mg/kg and lead ≤5 mg/kg. Solubility - insoluble in water and ethanol.

6.1.5 European Food Safety Authority

The EFSA Panel on Additives and Products or Substances used in Animal Feed (FEEDAP) issued a scientific opinion on the additive clinoptilolite of sedimentary origin that has by specification at least 80% clinoptilolite and a maximum of 20% clay minerals (EFSA, 2013). The main chemical composition and purity of the clinoptilolite reported in the scientific opinion (based on eight batches) is shown below.

> Clinoptilolite:
> Composition - SiO_2 (67%), Al_2O_3 (12%), CaO (3%), Na_2O (0.7%) and Fe_2O_3 (1.3%).
> Impurities - concentration of heavy metals (mg/kg): Cd (<0.04), Pb (<7.4), Hg (<0.005) and As (<1.4). Dioxins in four batches analyzed did not exceed 0.1 ng WHO-PCDD/F-PCB-TEQ/kg.
> Physical data - Laser diffraction analysis of one batch of the additive identified 90% (v/v) of the particles with a diameter of ≤186 µm, 50% with a diameter ≤72 µm and 31% with a diameter ≤50 µm. Laser diffraction analysis of three other batches identified 50% of the particles falling within the respirable fraction (≤8 µm) and 10% of the particles being ≤1 µm.

EFSA concluded that clinoptilolite is considered to be safe for all animal species when used as a binder/anticaking agent in organic animal feed, subject to the limit of 10,000 mg/kg (1%) of complete feeding stuff as specified in EU Regulation 651/2013 (EFSA, 2013; EU, 2013). Clinoptilolite is also listed under the category technological additive, functional group binder/anticaking agent, code 1g568 in the European Union Register of Feed Additives (EU, 2018).

6.1.6 Supportive data for clinoptilolite

As can be seen from the information reviewed above, data on the composition of the three aluminosilicate compounds listed as GRAS when used as anticaking agents is limited. Sodium aluminosilicate is relatively well described, and some information exists for aluminium calcium silicate, but not for sodium calcium aluminosilicate. Interestingly, composition data for sodium aluminosilicate presented in the SCOGS report was not included in the later FDA GRAS listing. To date, a description of the composition, purity and related parameters for the three aluminosilicate compounds does not exist (FDA, 2016a,b). A detailed description of the composition and purity of clinoptilolite authorized as an anticaking agent in the EU is given by EFSA (2013).

A common property of these anticaking agents is that they belong to the mineral class of aluminosilicates, which contain the same main components: SiO_2 and Al_2O_3. Clinoptilolite, as per its chemical composition meets this criterion of being an aluminosilicate anticaking agent. In this regard, the safety of adding clinoptilolite to the diets of food-producing animals is addressed in detail in the next section.

6.2 Feeding studies in target animals with clinoptilolite

Clinoptilolite has been used in animal nutrition since the mid-1960s (Mumpton and Fishmann, 1977). The literature contains numerous studies in which ruminant, swine and poultry diets were supplemented with this mineral substance as an additive (Ramos and Hernández, 1997; Papaioannou et al., 2005; Karamanlis et al., 2008, Pourliotis et al., 2012; Karatzia et al., 2013; Subramaniam and Kim, 2015). However, the adding of clinoptilolite to feed as a binder for mycotoxins or any use other than as an anticaking agent is not considered as being GRAS (IDALS, 1999; FDA, 2016c).

The scientific data and information presented in Sections 6.2.1 through 6.3.4 demonstrate that clinoptilolite can be safely fed to food-producing animals. Emphasis is given to studies where the amount of clinoptilolite added to diets was equal to or greater than the proposed GRAS allowable tolerance of 1%. The main study parameters reviewed are the number and type of animals used, length of the trial and whether adverse effects on performance or general health were observed. Any additional parameters that may have been concurrently measured or compared (statistically or otherwise) as part of a cited study are included only as relevant. As required by 21 CFR 570.250 we discuss also available studies on related substances or used at much higher concentrations though those are of limited evidence on the safety of the clinoptilolite subject to this notification when used at 1% in feed.

6.2.1 Ruminants

Cattle

McCollum and Galyean (1983) conducted a 148-day feedlot experiment with clinoptilolite. Forty-eight cross-bred steers (average body weight 304 kg) were randomly divided into three groups and fed a 70% sorghum diet with clinoptilolite substituted at 0, 1.25 and 2.5% of the diet dry matter. Because the steers had recently been removed from a grazing trial, they were allowed a 2-week period for environmental and dietary adaptation. Once the steers were on full feed, the experimental diets were offered, and the feeding trial commenced. During the experiment, fresh feed was offered once daily in amounts sufficient to allow free choice consumption. On days 1, 37, 66, 93 and 148, all steers were weighed, and fecal grab samples were collected from four randomly selected steers/pen. All steer weights were adjusted using a 4% shrink. The pH of fecal grab samples was determined, and the samples were frozen until completion of the trial. Steers were slaughtered at a commercial packing plant after 148 days on feed and yield of lean cuts and USDA quality grades were determined by a federal grader. No differences (P>0.05), were found at 1.25 % clinoptilolite (as compared to the controls) in dry matter intake, average daily gain, feed to gain ratio, carcass weight, dressing percentage, total digestion, rumen digestion and intestinal digestion.

Hutcheson (1984) administered three types of clinoptilolites to steer calves (average weight 229 kg) at levels of 3% comparing it to a control diet. One of the clinoptilolites (C20) had a purity of 88%, which is comparable to the notifier's product. The steers were maintained for 56 days in open pens equipped with individual feed monitoring devices and weighed at days 0, 7, 14, 28, and 56 of the study. Blood samples taken at these times were analyzed for packed cell volume; whole blood K and serum K, Na, inorganic P, Ca, Mg, Cu and Zn. At 56 days, average daily gains were similar for the C20 treatment (1.25 kg) and the control (1.32), with feed to gain ratios of 8.38 and 5.95, respectively. The authors claim that serum sodium was significantly lower after 56 days, and whole blood potassium and serum phosphorus increased significantly at 7 days for animals on the C20 diet. No differences were detected for the other blood and serum indexes measured. A closer inspection of the serum values reported by the authors in Table 5 of their publication questions their conclusion and interpretation: sodium levels at day 56 of the C20 group are 139.0 meq/L which is equal to the level of the control group at day 0. For potassium already at day 0 levels for C20 are significantly higher and show a much larger standard deviation indicating an unequal distribution between animals from both groups. The phosphorus values for the C20 group at day 7 are lower than the control group at day 28. Over the 56 days of the study for all studied blood parameters values fluctuate significantly and the claimed statistically significant changes are not treatment related if the whole data population is considered.

Katsoulos et al. (2005a) investigated whether the long-term supplementation of 1.25% and 2.5% clinoptilolite in the concentrate feed of dairy cows had any effect on their hematological parameters. Fifty-two clinically healthy Holstein cows were randomly assigned to one of three groups according to their age and parity. Group 1 (control, n = 18) received concentrate feed. Group 2 (n = 17) and Group 3 (n = 17) received concentrate feed supplemented with 1.25% and 2.5% clinoptilolite, respectively. The experiment started 30 days before the expected parturition and lasted up to the end of lactation. Blood samples from individual animals were collected just before the start of experiment, at the day of calving and at monthly intervals. All samples were tested for packed cell volume, hemoglobin and leukocyte count (white blood cell) values. Results showed that packed cell volume values and hemoglobin concentrations were unaffected by clinoptilolite supplementation (P>0.05), either among groups or for individual sampling. Likewise, no differences (P>0.05) in WBC were observed between groups. Overall, the results showed that the 1.25 and 2.5% supplementation of clinoptilolite had no adverse effect on the hematological parameters tested.

Katsoulos and co-workers also evaluated a wide range of other parameters as part of this same study. The results from these studies are summarized here. Katsoulos et al. (2005b) investigated whether the long-term supplementation in the concentrate feed of dairy cows receiving diets supplemented with 1.25 and 2.5% clinoptilolite had any effect on the serum concentrations of ß-carotene and vitamins A and E. Results showed that clinoptilolite supplementation at both levels had no effect ($P>0.05$) on serum concentration levels of the three parameters measured.

Katsoulos et al. (2005c) examined the effect of the percentage of clinoptilolite fed to the two groups of dairy cows on the incidence of parturient paresis and serum concentrations of total calcium (tCa), inorganic phosphorus (PO_4^{2-}), magnesium (Mg^{2+}), potassium (K^+) and sodium (Na^+). Results showed that the incidence of parturient paresis in cows receiving 2.5% clinoptilolite was lower ($P<0.05$ than for the control animals. However, serum concentrations of Ca, Phosphate, Mg, K and Na were not different ($P>0.05$) in cows receiving either 1.25 or 2.5% clinoptilolite over a 30-day days experimental period.

Katsoulos et al. (2005d) investigated the effect of feeding two levels (1.25% and 2.5%) of clinoptilolite on serum copper, zinc and iron concentrations. Fifty-two clinically healthy Holstein cows were randomly assigned to one of three groups according to their age and parity. The first group (group A) comprised 17 cows fed a ration supplemented with 1.25% clinoptilolite, the second group (group B) comprised also 17 cows was given a ration with 2.5% clinoptilolite, and the third group (group C, the control), comprised 18 cows fed the basal ration that did not contain any clinoptilolite. The experiment started when the cows entered the fourth week before the expected parturition and lasted until the end of lactation. All cows were fed the above concentrates during the entire experimental period. Blood samples were collected from each animal at the starting day of the experiment, at the day of calving, and at monthly intervals thereafter. All samples were tested for serum Cu, Zn, and Fe concentrations. The results showed that the 1.25 and 2.5% supplementation of clinoptilolite did not have any adverse effects on serum concentrations of Cu, Zn, and Fe.

Katsoulos et al. (2006) analyzed blood samples monthly for serum glucose, ketone bodies, liver enzymes, blood urea nitrogen (BUN) and total proteins. The milk yield of each cow was also recorded monthly. The cows receiving 2.5% clinoptilolite had significantly fewer ($P<0.05$) cases of clinical ketosis during the first month after calving and a higher total milk yield. Feeding the cows with clinoptilolite for a long period had no apparent adverse effects on their liver function and did not significantly affect ($P>0.05$) the concentrations of glucose, ketone bodies, BUN and total proteins in their serum.

Bosi et al. (2002) assessed the effect of a clinoptilolite addition of 1 % on production and milk composition of dairy cows, on rumen fluid composition, and on the content of certain minerals in the blood over 76 days. Thirty-two lactating Holstein cows (142 days average lactation length) were blocked according to milk production, parity, and days of lactation and fed a control diet based on corn and alfalfa silages, hay and concentrates, or the control diet plus clinoptilolite (200 g/day). The clinoptilolite supplementation had no significant effect on milk yield, milk protein contents, fat contents and somatic cell count. The dietary addition of clinoptilolite did not change pH, ammonia content and VFA molar percentages in the rumen. No dietary effect on mineral contents of blood plasma (Na, K, Zn, and Ca) was observed. The test material used in this study had a clinoptilolite content of 45% which is significantly lower than specified in the present notification.

Enemark et al. (2003) monitored serum and urine biochemical changes in dairy cows during and after oral administration of a synthetic sodium aluminum-silicate (zeolite A) with a specified calcium binding capacity of > 110 mg per g zeolite (at pH 11). The animals received in addition to a grass silage ration as part of a pellet daily 709 g of zeolite from day 8 to day 14 of the study The following blood and urine parameters were investigated before, during, and after zeolite administration: blood gas, serum calcium, serum inorganic phosphate, serum magnesium, serum sodium, urine pH, urine calcium, urine inorganic phosphate, urine magnesium, vitamin D, parathyroid hormone (PTH), and urine pyridonoline and deoxypyridonoline. A slight decrease in serum Ca and in renal excretion of calcium was observed in the experimental group at initiation of supplementation, whereas an increment in these parameters was recorded after withdrawal of zeolite supplementation. It was assumed, that zeolite caused a reduction in the availability of dietary calcium during supplementation, which possibly elicited an activation of calcium mobilization. The influence of zeolite on calcium homeostasis was not evident from monitoring serum concentration of calcium regulating hormones (PTH, 1,25(OH)D3, 25(OH)VitD) or renal excretion of markers of bone resorption. Enhanced active intestinal calcium absorption and bone resorption was therefore considered insignificant in the calcium mobilization under the conditions of the experiment. Serum concentration and fractional excretion of magnesium and phosphate during supplementation were significantly lower in treated animals than in the control group. The influence of zeolite on phosphorus and magnesium was presumed to result from a combination of interference of zeolite with intestinal absorption and a marginal dietary supplementation of these minerals. The test material used in this study was of synthetic origin selected for its calcium binding properties; this type of zeolite is not comparable to the clinoptilolite subject to this notification. Furthermore, the daily amount fed (about 700 g zeolite) corresponds to levels in feed between 5 to 10% (depending on the daily feed intake of dry cows). The study results show that synthetic sodium aluminum silicate of different composition may affect calcium, magnesium, and phosphate homeostasis if administered at high daily doses, but such effects will not be present when adding clinoptilolite at 1% to the feed.

Mohri et al. (2008a) studied the effect of clinoptilolite supplementation (2%) to colostrum (for 48hr) and to colostrum and milk meals for 14 days in neonatal calves. Blood samples were taken 12 hours after birth and weekly until the end of the experiment after 42 days. Clinoptilolite supplementation for 14 days had significant effect ($P<0.05$) on the values of hematocrit, red cell count, hemoglobin, monocyte, and albumin. At the 48 hours supplementation mean corpuscular value decreased. No significant difference was seen for other measured parameters, performance, and health between trial groups. In a separate publication the same authors (Mohri et al.., 2008b) reported effects on some serum minerals. The described study protocol was the same, levels of calcium, phosphorus, magnesium, iron, sodium, and potassium were determined in the serum. The supplementation had significant effect on the concentrations of calcium, phosphorus, sodium, and iron. The concentrations of iron were significantly higher after 14 days of supplementation, and calcium and sodium concentrations were significantly higher in serum of clinoptilolite-treated than control calves. The concentrations of phosphorus were significantly lower in test groups than control group. Potassium and magnesium concentrations were not affected by clinoptilolite supplementation. The use of clinoptilolite at 2% in colostrum or milk replacer for neonatal calves does not represent an anticaking agent use. The observed effects were statistically significant, but small if compared to the ranges for these parameters reported elsewhere (Bouda and Jagos, 1984; Brun-Hansen et al., 2006; Klinkon and Jezek, 2012) and not of adverse nature. The results of this study where clinoptilolite was used at 2% in liquid feed do not have impact on the safety of clinoptilolite when used as anticaking agent in dry feed materials at levels of 1%.

Step et al. (2008) aimed at identifying any adverse effects on health or performance in young dairy calves fed clinoptilolite mixed with milk replacer. Twenty-six male Holstein calves (1 to 7 days old) were fed milk replacer with no clinoptilolite (n = 8), 0.5% clinoptilolite (n = 9), or 2% clinoptilolite (n = 9) for 28 days; each calf consumed approximately 12% of its body weight per day (based on the replacer solids in the milk replacer mixture). For each calf, subjective health assessments, weight and rectal temperature measurements, and complete blood cell count and serum biochemical analyses were performed at intervals. All calves underwent necropsy. Body weight and average daily gain did not differ among treatment groups. The percentage of monocytes and serum total protein concentration in the low-dosage group were higher than values in the control and high-dosage groups. Compared with values for either clinoptilolite-treated group, blood urea nitrogen concentration was greater in the control group. Serum globulin concentration differed significantly among groups (2.77, 2.50, and 2.36 g/dL in the low-dosage, control, and high-dosage groups, respectively). At necropsy, gross lesions associated with clinoptilolite treatment were not detected in any of the calves. The authors concluded that even under stressful conditions, clinoptilolite fed at 0.5 % or 2.0% did not affect the performance of dairy calves and had no negative effect on white blood cell counts and blood metabolite concentrations and enzyme activities. Clinoptilolite ingestion was not associated with treatment-specific gross changes. The noted changes in blood urea nitrogen and serum globulin are statistically significant, but small if compared to the ranges for these parameters reported elsewhere (Bouda and Jagos, 1984; Brun-Hansen et al., 2006; Klinkon and Jezek, 2012) and not of adverse nature.

Zarcula et al. (2010) and Pourliotis et al. (2012) studied the effects of clinoptilolite on diarrhea in neonatal calves, an endpoint that is not covered by this submission. Zarcula et al. (2010) added 0.5% and 2% to colostrum or milk replacer, and Pourliotis et al. (2012) administered doses of 1 g/kg body weight per day. Both studies did not report any adverse effects and support the general safety profile of clinoptilolite when used in neonatal calves.

Sweeney et al. (1984) studied in Holstein steers and heifers in a cross-over design the effect of 5% clinoptilolite on diets with different protein solubility (low vs. high) fed for 112 days. Blood and rumen parameters, and fecal and urinary parameters were measured and determined at regular intervals during the whole duration of the study. Although a tendency for increased feed intake was found, neither daily weight gain nor daily intake was significantly affected by clinoptilolite in both types of diets. A significant clinoptilolite-protein solubility interaction was observed for apparent digestibility of protein, organic matter, and acid detergent fiber. Clinoptilolite caused a shift in the ration of acetate:propionate from 2.75 to 2.96. In the blood urea nitrogen increased with the low-solubility diet and decreased in the high-solubility diet, and potassium decreased in both. Overall in this study the use of two diets different in protein solubility is a confounding factor. As the only investigated level is five times higher than the proposed use as anticaking agent of 1%, the study results do not question the safe use of clinoptilolite as discussed in this notice.

Karatzia et al. (2011) reported results from a study in dairy cows where animals received 200 g of clinoptilolite per day. The test material was offered together with a concentrate (4 kg), corn silage (25 kg) and molasses (2 kg). The aim of this study was to investigate whether the dietary inclusion of 200 g of clinoptilolite has any effect on the blood serum concentrations of aluminum and inorganic phosphorus, as well as on the ruminal pH and the ruminal concentrations of aluminum and of certain volatile fatty acids. The aluminum concentrations in rumen fluid and blood serum were not affected by clinoptilolite supplementation throughout the experimental period which indicates that clinoptilolite was stable at the acidic pH of the gastrointestinal tract. Concentration of soluble phosphorus in the rumen fluid was not significantly affected by clinoptilolite supplementation. Clinoptilolite significantly increased the pH of the rumen fluid, an increase that was attributed to the buffer effects of clinoptilolite when added to acidic or basic aqueous solutions. The total volatile fatty acid concentration was not significantly affected by clinoptilolite supplementation, but the molar proportions of the volatile fatty acids evaluated were significantly affected by clinoptilolite. Acetate was increased while propionate and valerate were decreased.

The authors' findings contrast with results obtained by other authors; however, it is justified to question that the observed changes were negative effects. It is generally acknowledged that an increase in rumen pH and acetate, and a decrease in propionate are beneficial. The levels achieved in this study are those you would expect in healthy animals and hence are not an evidence of clinoptilolite being unsafe for dairy cows (Krause and Oetzel, 2006; Enemark, 2008; Abdela, 2016)[1].

Sheep and goats

Pond et al. (1984) carried out a study with 54 growing male lambs to determine the effect of dietary clinoptilolite on growth, feed utilization and concentrations of several nitrogen and ionic constituents in blood plasma and of selected elements in body tissues. The treatments consisted of a basal (low protein) diet containing no nitrogen supplement, plus vitamins and minerals, and a similar diet fortified with urea as a nonprotein nitrogen source or with soybean meal as an intact protein source added to the diet isonitrogenously with urea, all fed for 9 weeks. Clinoptilolite was added to each of these three diets at 3% at the expense of corn. Mean daily weight gain was depressed during the first 4 weeks and at 9 weeks in lambs fed diets containing corn or urea when clinoptilolite was added ($P<0.05$). However,

[1] Standard Text books that discuss the need of avoiding low rumen pH and low acetate/propionate ratios are among others: "Merck Veterinary Manual", Dukes' Physiology of Domestic Animals" and "Veterinary medicine: A textbook of the diseases of cattle, sheep, goats, pigs and horses."

when clinoptilolite was added to the corn-soybean meal diet there was no growth depression for either period. Clinoptilolite did not affect plasma concentrations of protein, Ca, P, Mg, Na or K at 9 weeks. The pH of the gastrointestinal tract contents was not affected, except for an increase in pH of caecal contents due to clinoptilolite, a mild effect if compared with the fluctuations the study authors report for the various measurements made at different segments of the GI tract There was no evidence of changes in liver, kidney, testes and muscle concentrations of major or minor elements in lambs fed clinoptilolite. The test material used in this study was of about 60% purity which is less than the product specified in Part 2 of this Notice. However, the results support the general safety of clinoptilolite as no adverse effects were reported at 3% dietary inclusion.

Kovac et al. (1995) studied the use of clinoptilolite-rich tuff (about 60% clinoptilolite) to alleviate organophosphate intoxication in sheep. The administration in this study was by esophageal intubation directly into the stomach, the applied single dose was 2 g/kg bw. The results suggest that clinoptilolite-rich tuff administrated orally 20 min before application of an organophosphate in sheep appears to provide partial protection against the toxin in low dosages. It should be noted that the mode of administration does not allow the use of this study for a feed safety evaluation. The use of the study is further limited by the fact that animals received the pesticide at toxic concentrations.

Deligiannis et al. (2005) assessed the effect of feeding clinoptilolite (with a content of 88%) on food intake and performance of growing lambs infected or not with gastrointestinal nematodes. The experiment was carried out over a period of 3 months. Twenty-four lambs of the indigenous Greek dairy breed Karagouniko were used. They had been weaned at 6 weeks of age. Lambs were given free and continuous access to a nutritionally non-limiting pelleted concentrate that was either a basal diet or a diet supplemented with clinoptilolite at a level of 3%. A 2 x2 factorial design consisting of both feeding treatments and two levels of parasitic status, infected and uninfected was used. The infected animals received 15000 L_3 (larvae) of GI nematodes. Fecal samples were obtained directly from the rectum of each individual animal. Sixty-two days later, all lambs were slaughtered and their abomasa with intestines were removed in order to recover the adult worms. Lambs whether infected or not, without clinoptilolite in the feed did not increase their feed intake to the same extent as those receiving clinoptilolite. There was also a significant difference in the live weight of lambs at slaughter, with lambs that received zeolite being the heaviest. Average growth rates were significantly different between lambs of different groups; lambs of groups with clinoptilolite were growing to a faster rate compared with those without. However, there were no significant differences in carcass characteristics between lambs of the four groups. The fecal egg counts were significantly reduced in infected lambs receiving clinoptilolite; the latter was also the case for worm burdens assessed on the day of slaughter. In this study at 3% inclusion rate were not observed negative effects on growth parameters of lambs.

Ruiz-Barrera et al. (2006) determined the effect of four levels of clinoptilolite on the digestibility and nutrient intake of sheep fed with alfalfa hay and concentrate. Four fistulated male sheep with an average 32.55 (±1.43) kg of liveweight were allocated to four treatments in a Latin square design: 0% clinoptilolite, 1.5% clinoptilolite, 3% clinoptilolite and 4.5% clinoptilolite. Results for apparent dry matter digestibility, dry matter intake, organic matter intake, acid detergent fiber intake, neutral detergent fiber intake and crude protein intake the data showed no statistical differences. However, a significant quadratic effect was observed for digestible intake of acid detergent fiber with values of 72.0 (0%), 94.4 (at 1.5%), 98.6 (at 3 %) and 87.3 (at 4.5%) g/animal/day. In this study no side effects were reported, however, the authors recognize the limitations of their study with respect to the sample size.

Katsoulos et al. (2009) determined the effect of dietary supplementation with clinoptilolite (89% purity) on health and production as well as serum concentrations of fat-soluble vitamins, macro elements and trace elements, and activities of hepatic enzymes in dairy goats. Goats were randomly assigned to a clinoptilolite group (n=36) that received concentrate feed, of which 2.5% contained clinoptilolite, the control group (n=36) received unsupplemented feed. The experiment began 8 weeks before parturition and continued to the beginning of the next nonlactating period (280 days of lactation). At the day of parturition, kids were weighed. Milk yields were recorded at day 60 of lactation and thereafter at monthly intervals. Milk percentages of fat, protein, and lactose and somatic cell count were evaluated at the same points. Blood samples were obtained at the beginning of the experiment, the day of parturition, and thereafter at monthly intervals to measure serum concentrations of fat-soluble vitamins, macro elements and trace elements, and activities of hepatic enzymes. Birth weights of triplets and quadruplets were significantly higher in clinoptilolite-treated goats versus control goats. Milk fat percentage was significantly higher and somatic cell count was significantly lower in clinoptilolite-treated goats, compared with respective values in control goats. No changes in serum concentrations of any variable were detected. The authors concluded that clinoptilolite supplementation of concentrate feed at 2.5% improved milk fat percentage in dairy goats, without adverse effects on the serum variables evaluated. The clinoptilolite used in this study was of similar quality as described in Part 2 of this Notice.

Norouzian et al. (2010) examined the effects of feeding clinoptilolite on hematology, performance, and health of 30 newborn Balouchi lambs allocated to three groups: one with control basal diet and two basal diet plus 1.5 and 3% clinoptilolite, respectively, for 6 weeks (3 weeks before and 3 weeks after weaning). Blood samples were taken from all lambs, at the time when the animals were allocated to the experimental diet and at the end of each week of experiment, and analyzed for hematology, plasma fibrinogen, and total protein. Performance and health of all lambs were measured. Fecal consistency score and diarrhea severity were evaluated. There was no difference between lambs in case of hematological parameters. Lambs fecal consistency score and severity of diarrhea were lowest ($P<0.05$) for both groups of clinoptilolite-supplemented lambs and highest for control animals. Feed conservation ratio and dry matter intake were similar between the groups of lambs fed the different diets, but average daily gain of lambs differed significantly ($P<0.05$) and was higher in animals supplemented with 1.5 % clinoptilolite. It was concluded that addition of 3% clinoptilolite to starter diet of newborn lamb can reduce incidence and severity of diarrhea, although its effect on hematology and performance was negligible.

Alcala-Canto et al. (2011) evaluated the effects of supplementation with clinoptilolite in a controlled and randomized field study conducted on a sheep farm with a known history of coccidiosis. Two groups of 6 pregnant ewes each, with three replicates per group were included in the study. Sheep received a basal diet or a clinoptilolite (1.25%) diet. Clinoptilolite was fed to the experimental group for a mean of 72 days (30 days before lambing through 42 days after lambing) and examination of fecal samples was performed every seven days in ewes, as well as on days 12, 14, 21, 28, 35 and 42 in their lambs. The assessment was based mainly on total oocyst excretion of *Eimeria* oocysts in ewes and their offspring, and on morphology of *Eimeria* oocysts. On day 42, efficacy reached a significant ($P<<0.05$) reduction of 97% in oocyst output in supplementation. Of relevance for the safety of clinoptilolite are the reproductive parameters: There was no negative effect of supplementation with clinoptilolite on ewe weight gain, number of lambs born and newborn lamb body weight (the supplementation rather improved those, possibly due to reduced *Eimeria* infection). The study authors do not discuss the purity of the clinoptilolite used; the results support the supplementation safety of clinoptilolite at 1.25% in diets of reproducing sheep.

Toprak et al. (2016) conducted a study to determine the effects of the addition of micronized clinoptilolite on the fattening performance, blood parameters, fecal ash and nitrogen levels of lambs fed concentrate feeds intensively. For two months 25 four-month-old Merino x Ile de France crossbred male lambs (21.1 ± 1.32 kg live weight) were fed 100 g alfalfa hay and a mixed concentrate diet containing 0%, 1%, 2% or 3% additional clinoptilolite. At the end of the study, bodyweight gain and feed consumption were not affected by the treatments. Similarly, the addition of up to 2% clinoptilolite to the diet did not affect slaughter weight, hot carcass or cold carcass weights, but they decreased at 3% inclusion. No differences were observed between the groups in terms of blood urea nitrogen, plasma glucose, serum creatinine, triglyceride, sodium, potassium and chlorine concentration. However, serum total protein, calcium and phosphorus concentrations were affected by clinoptilolite supplementation of 2% or 3%. The addition of clinoptilolite to the ration did not affect the fecal dry matter content and total nitrogen level, yet it increased the ash content of the feces. Consequently, it was demonstrated that the addition of up to 2% clinoptilolite to lamb grower feed does not have a negative impact on performance and carcass yield of the animals, but affects serum total protein, calcium and phosphorus concentrations. No effects were seen at a 1% inclusion rate.

6.2.2 Swine

Nestorov (1984) investigated the effects of a clinoptilolite (65% purity) when fed to male pigs over a growth period from 10 to 105 kg body weight. In the treated animals' diet 4% carbohydrates were replaced by 4% of clinoptilolite. During the finishing period, between 30 and 103.3 kg of weight, mean daily weight gains were 0.698 and 0.687 kg and feed consumption was 3.1 and 3.02 kg for the control and experimental groups, respectively. At slaughter, a significant difference (P <0.01) was found in carcass lean and fats for the two groups. The experimental animals had a 9% greater lean content and a 16% lower separable fat (subcutaneous and interior fats) content than the control animals. In a second large-scale field experiment, two groups of Camborough pigs were equalized on the basis of live weight and age and fed a basal control diet or a similar diet in which 4% of the maize was replaced by clinoptilolite. Thus, during the growth period from 30 to 60 kg of live weight, the control-group animals received 14.15% crude protein and 4012 kcal of metabolizable energy per kilogram, and the experimental animals received 13.15% crude protein and 3890 kcal of metabolizable energy per kilogram. For the period between 60 and 105 kg of live weight, the groups received 12.33 and 11.8% crude protein and 3624 and 3328 kcal metabolizable energy per kilogram, respectively. For the entire finishing period, mean daily weight gains of 0.471 kg and 0.461 kg were obtained for the experimental and control groups, respectively. Feed consumption was 4.598 kg/kg weight gain for the experimental animals. In growing pigs fed these diets, significantly higher levels of blood glucose and alkaline phosphatase and lower levels of blood lipids were found in the mesenteric blood of the experimental animals. Blood serum monoamino oxidase activity was significantly higher in the control pigs, which was related to the higher levels of lipids in the serum. It should be noted that several aspects of the study limit its relevance for this notice: (a) the purity of the clinoptilolite is lower; (b) the diets differed significantly, as carbohydrates were replaced without compensation; (c) the inclusion level is four times higher than proposed in present notification; (d) no data for blood parameters are given which does not allow to assess retrospectively whether the changes are within or outside of normal ranges (i.e. adverse) and related to clinoptilolite or the different diets.

Shurson et al. (1984) carried out several trials in which growing pigs were fed diets containing various levels of clinoptilolite. In one growth trial, two groups of 18 crossbred pigs (averaging 25 kg body weight) were assigned to diets containing 0 or 5% clinoptilolite for 6 weeks growing phase trial. It was found that average daily gain (ADG), average daily feed intake (ADFI) and F/G ratio were unaffected by clinoptilolite supplementation. A second growth trial utilized the same crossbred pigs (averaged 65 kg body weight) assigned to diets containing 0 or 5% clinoptilolite for 8 weeks finishing phase trial. No effect was observed for ADG, ADFI or metabolizable energy (ME) utilization with the clinoptilolite diet, although the F/G ratio increased ($P<0.02$). In a nutrient balance trial, 16 crossbred pigs (averaging 7 kg body weight) were fed diets (4 pigs per diet) containing 0, 2.5, 5.0 or 7.5% clinoptilolite. Digestible energy, ME, N-corrected ME and ME corrected for N-balance and clinoptilolite levels were linearly reduced ($P<0.01$) as increasing amounts of clinoptilolite were fed. Daily fecal-N increased, and apparent digestibility of N was linearly reduced by feeding increasing amounts of clinoptilolite. Likewise, phosphorus retention was linearly reduced ($P<0.05$), but Ca, Mg, Na, K, Fe and Zn retention were not linearly affected. The authors stated that this indicated the stability of clinoptilolite in the acid environment of the stomach.

Sardi et al. (2002) carried out two trials in which pigs were fed diets containing clinoptilolite. In the first trial (growing-finishing) 40 pigs (average 55 kg body weight) were homogeneously allocated to two groups (4 replicates of 5 animals each) and fed either a standard diet or the same diet supplemented with 2% clinoptilolite. Pigs were slaughtered at about 160 kg body weight. Blood samples were taken to determine BUN. In the second trial (piglets from birth to post-weaning) a total of 116 piglets from 12 litters were used. Sows were homogeneously chosen with respect to farrowing order (second parity-dams) and genetics (Duroc x Large White). Piglets were weighed at birth. Litters were equalized to assign to each sow the same number of piglets of the same sex. Litters were allotted to two groups: the control group in which the 6 litters received feed without clinoptilolite and the experimental group in which the remaining 6 litters received the control diet with 2% clinoptilolite. Animals were fed until attaining 33 kg body weight. The ADG, ADFI and health of the pigs were regularly recorded in both trials. Results from both trials showed that the inclusion of clinoptilolite did not affect growing performance. Animals fed the clinoptilolite diet showed a significant (P<0.05) improvement of fecal DM content. It was observed at slaughtering that the dietary inclusion of clinoptilolite resulted in a trend towards an improvement in yield of lean cuts and in an increase (P<0.05) of the ratio between lean and fat cuts. The authors stated that clinoptilolite did not impair pig growing performances, resulted in a higher dry matter content of feces and improved carcass quality of heavy pigs, particularly in regard to yield of lean cuts and ratio of lean/fat cuts.

Papaioannou et al. (2002) conducted a study to evaluate, under field conditions, the effect of the long-term dietary use of a natural zeolite (clinoptilolite) and antibiotics (chlortetracycline) on the concentrations of certain vitamins (vitamin A and vitamin E) and minerals (K, Na, P, Ca, Mg, Cu and Zn) in blood and body tissues of weanling Large White x Landrace sows. Twenty-four sows were assigned to two main experimental groups and four subgroups, depending on the presence or absence of clinoptilolite and chlortetracycline in their feed, respectively. Clinoptilolite was provided to the sows from weaning, during the service, gestation and lactation periods and up to the date of the next service, while chlortetracycline was administered for a 2-week period post-service, as well as for a 2-week period following the allocation of the sows in the farrowing house, around 5 days prior to the expected parturition. Blood samples were collected at the starting day of the trial, on the thirtieth and ninetieth day of each pregnancy, and on the day of each parturition and on the day of each weaning. Furthermore, 20 sows were similarly distributed in the same experimental groups and subgroups, and at the end of the trial were slaughtered and liver and kidney samples were collected for biochemical analysis. Neither clinoptilolite nor chlortetracycline supplementation of the diets had any significant effect on vitamins' and minerals' uptake and their distribution in the body, since there was no alteration in their blood serum and liver/kidney concentrations. Furthermore, no clinoptilolite x chlortetracycline interaction was noticed. It was concluded that dietary supplementation of clinoptilolite during pregnancy and lactation is not associated with any adverse effect on vitamin (A and E) and mineral (K, Na, P, Ca, Mg, Cu, and Zn) uptake and/or distribution in sows which are on diets adequately fortified with these vitamins and minerals.

Fokas et al. (2004) carried out a study to measure the retention coefficient of lead (Pb) from clinoptilolite in a balance trial with growing pigs. Twelve weaned pigs of 45 days of age were divided into two equal groups. Group 1 was fed a control (C) diet and Group 2 (experimental, E) the same diet supplemented with 2% clinoptilolite. The Pb content of the clinoptilolite used was 46 ppm, and the dietary contents were 1.1 and 2.1 ppm for diets C and E, respectively. Feed intake, growth rate and feed conversion efficiency, as well as digestibility and nitrogen (N) retention were measured to investigate any effects of the dietary inclusion of clinoptilolite in pigs. Pb concentration was measured in whole blood and edible parts of the carcass (muscles, liver, heart and kidneys), and Pb retention coefficient was determined. The results showed that digestibility did not differ between diets C and E, apart from that of ether extract, which was lower ($P < 0.01$) for the C diet, whereas, N retention was higher in pigs of group E. No significant differences were observed for feed intake, growth rate and feed efficiency over the 52-day feeding period. The inclusion of clinoptilolite in the diet of Group E pigs did not result in a significant increase in the Pb concentration in the whole blood and the edible tissues, except for liver (P <0.001). However, the Pb retention coefficient between diets was not changed significantly.

Alexopoulos et al. (2007) studied the effect of the long-term dietary use of a clinoptilolite-rich tuff on certain biochemical and hematological parameters in pigs. Forty-eight healthy piglets (24 females and 24 males) were used in the study, equally allocated in two experimental groups, depending on the inclusion or not of clinoptilolite in their feed. Clinoptilolite was incorporated in their feed at the inclusion rate of 2% and was administered continuously from weaning to slaughter. All pigs were individually weighed upon commencement of the study at 25(±3) days of age and subsequently at the end of the weaning, growing and fattening stages; 70(±3), 112(±3) and 161(±3) days of age, respectively, and blood sample collections were performed for analyses. Clinoptilolite ingestion was well tolerated and resulted in a higher body weight gain during the weaning stage, as well as during the whole monitoring period ($P<0.05$). The diet had no effect on serum K, Na, Ca, P, total protein, albumin and total bilirubin concentrations throughout the study ($P>0.05$). Furthermore, no significant alteration was noticed concerning the hematocrit, leucocyte count and hemoglobin concentration ($P>0.05$). Conversely, the dietary inclusion of clinoptilolite resulted in a lower serum urea-N and cholesterol concentrations in all blood samplings apart from the initial one ($P<0.05$). Likewise, the clinoptilolite supplemented group demonstrated an elevated serum glucose concentration at the blood samplings performed on 112(±3) and 161(±3) days of age and a lower AST activity at the latter ($P<0.05$). In the context of this study, the long-term dietary use of clinoptilolite at the inclusion rate of 2% appeared not to affect adversely the health status of treated animals in terms of undesirable changes of their biochemical and hematological profiles.

Prvulovic et al. (2007) carried out a study to determine the effects of clinoptilolite on growth performance and selected blood serum biochemical components in Landrace × Yorkshire pigs. Sixty animals of both sexes were divided into two groups and fed either a basal diet without clinoptilolite or the same diet supplemented with 5% clinoptilolite. Feed and water were available ad libitum. Individual live weights were recorded on days 45, 90 and 135 of the experiment. Feed consumption was recorded weekly. Blood was drawn from the anterior vena cava of each pig at day 135 for the determination of serum biochemical parameters. During the first 90 days of the experiment, pigs from the clinoptilolite group had higher body weight gain ($P<0.05$) compared with the control group, but growth parameters in the finishing phase were significantly lower ($P<0.05$). This decrease was attributed to possible effects of the concentration, purity and type of clinoptilolite used, as well as the growth phase of animals. Blood serum biochemical parameters from all experimental pigs were reported as being generally within the normal range, although higher ($P<0.05$) triglyceride concentration, lower total cholesterol concentration and increased activity of aspartate amino transferase were recorded in the serum of the clinoptilolite supplemented group. Dietary addition of clinoptilolite had no adverse effects on other serum biochemical parameters and did not affect the normal physiological homeostasis of the animals.

The literature contains several additional studies that support the safety of adding 1% or more clinoptilolite to swine diets. For example, see the reviews by Pond et al. (1989), Papaioannou et al. (2005), Alexopoulos et al. (2007) and Subramaniam and Kim (2015).

In view of the numerous sources of clinoptilolite and their wide range of applications, studies available from the literature are sometimes quoted as presenting evidence for hazards that may raise concerns when clinoptilolite is added to animal diets. However, as demonstrated by the example of EFSA (2013) described below, clarification of such concerns is needed before applying the evidence to other studies.

According to EFSA which referenced the study from Pond et al. (1988), "*some changes in the mineral concentration of the kidney of pigs were found*". This study investigates co-administration of clinoptilolite with higher doses of copper. It seems that EFSA did not consider the second publication by Pond et al. (1989) where the authors assessed further the tissue mineral element content in swine. In this study fattening pigs received diets low and high in calcium with low and high content in iron, plus 0 or 2 % clinoptilolite. The authors determined in liver and kidney the levels of sulphur, phosphorus, potassium, sodium, magnesium, iron, aluminium, copper and zinc. There was no effect of clinoptilolite on liver concentration of any mineral element measured. They noted small shifts in the kidney level of potassium, magnesium and copper, however, those were of no consequence to overall animal health or tissue or cellular integrity, since no pathological lesions or clinical signs of aberrations in mineral element metabolism were evident. The authors concluded that the addition of 2% clinoptilolite to corn-soybean meal diets fed continuously to growing-finishing pigs for 84 days was not associated with adverse effects on tissue mineral element concentrations even when dietary calcium and iron concentrations are altered. The several observed changes in tissue mineral concentrations in response to dietary clinoptilolite do, however, suggest that dietary mineral element composition should receive special attention when clinoptilolite is added to the diet. It is worthwhile to note that Pond and Lee (1984)[2] studied incorporation levels of 5% clinoptilolite, but found no effects on blood plasma traits that were related to the addition of clinoptilolite; blood urea increased significantly only at the co-administration of ammonium carbonate, clinoptilolite alone did not change blood urea nitrogen as compared to controls.

The study of Poulsen and Oksbjerg (1995) investigated effects of 0 and 3% clinoptilolite on protein-poor and -rich diet. The feed was not adjusted to the reduced energy content in the clinoptilolite group and, as pigs did not compensate for this effect by increasing feed intake, daily weight gain was slightly reduced. Additionally, the authors state that protein retention was not influenced significantly. The clinoptilolite used in the study contained less clinoptilolite (70%) than the clinoptilolite of this notice or the one authorized in the EU. Test material and inclusion rate also differ from our notification, and the observations made in the study are explained by the study design. Its results do not affect GRAS conclusion proposed in this notification.

6.2.3 Poultry

Olver (1989) carried out a study with 120 single combed 6-week-old pullets of three strains (20 hens per treatment) that were fed on a diet containing 160 g protein/kg with or without 5% clinoptilolite. Sterile river sand replaced clinoptilolite in the control diet in order to keep the diets isoenergetic. The hens were individually caged in a naturally-ventilated laying house and fed on one of the two diets for ten 28-day periods. No significant dietary effects between treatments were observed with respect to body weight, age at first egg, egg weight, Haugh scores or food intake/hen. Significant dietary effects in favor of clinoptilolite feeding were noticed with the number of eggs laid per hen, shell thickness, efficiency of food utilization, droppings moisture content and mortality. Significant differences between strains were observed with respect to all measurements taken, except food intake/hen/day.

Öztürk et al. (1998) conducted an experiment to study the effects of clinoptilolite on the performance of laying hens. A total of one hundred and eighty 37-week-old Babcock B-300 layers with similar egg production capabilities and live weight were divided into 5 treatment groups (12 replicates and 36 hens per treatment) and fed a diet containing 0, 2, 4, 6 and 8% clinoptilolite over a 112-day days experimental period. All feeding programs were isocaloric and isonitrogenous. No significant dietary effects were observed in terms of body weight, feed consumption, feed efficiency ratio, number of eggs laid per hen, shell thickness, mortality or other criteria of egg quality ($P>0.05$). Significant dietary effects of clinoptilolite feeding were observed in the form of a decrease in fecal moisture content ($P<0.05$).

Bintas et al. (2007) conducted a study to investigate the effect of dietary Ca (recommended and below recommended levels) and clinoptilolite (0 and 0.8%) on growth, plasma, tibia and feces in chickens from 1 to 42 days of age. Zeolite supplementation did not affect overall body weight gain, feed intake (FI) or feed conversion ratio (FCR) of broiler chickens ($P>0.05$). Overall mortality of clinoptilolite-fed chickens was lower than in untreated ones ($P<0.01$). Reduction of dietary Ca of approximately 10 to 18% decreased ($P<0.05$) body weight at 14 and 42 days of age in association with reduced FI, but overall FCR was unchanged. Serum protein and sodium constituents were reduced in birds fed clinoptilolite ($P<0.05$), but no significant effect in was observed in serum P, Mg, and Cl concentrations. Decreasing dietary Ca level increased ($P<0.01$) serum, total protein and glucose concentrations, but decreased Ca level. Zeolite decreased bone ash in birds fed a Ca-deficient diet while increased fecal excretion of ash, Ca, P and aluminum. However, clinoptilolite increased tibia weight ($P<0.05$) and thickness ($P<0.01$). No

[2] This paper is referenced sometimes as Pond and Yen (1984). however, the book *Zeo-agriculture: Use of Natural Zeolites in Agriculture and Aquaculture*. Westview Press, Inc. Boulder, CO, edited by WG Pond and FA Mumpton, list the authors of the paper as WG Pond and JT Lee. No paper published by Pond and Yen (1984) could be identified.

significant response (P>0.05) in relative weight and gross lesion scores of liver or footpad lesion scores was found related to changes in dietary regimes.

Eleroglu et al. (2011) investigated the effects of differing amounts of natural clinoptilolite + mordenite type of zeolite (Ca-zeolite) on bone and some blood parameters. A total of 240-day-old Ross 308 broiler chicks were assigned to four treatments with three replicates, each containing 20-day-old chicks of mixed sex. Ca-zeolite was added in the broiler diets at levels of 0 g/kg, 10 g/kg, 30 g/kg, and 50 g/kg. Stocking density was 14 broilers/m2. During the six-week trial, blood parameters and bone characteristics were monitored. The inclusion of Ca-zeolite, at various levels, did not have any significant effect on the concentration of blood serum biochemical parameters; serum glucose, cholesterol, total protein, uric acid; concentrations of Ca, P, Na, K, Cl, and on tibial bone characteristics (tibia weight, ash, volume, specific gravity, and Ca and P contents) in the two sexes and mixed-sex between the groups at 21 and 42 days of age.

Kralik et al. (2006) determined the effects of a preparation containing tribomechanically activated clinoptilolite on the egg quality and on the biochemical indicators in serum of laying hens. The experiment was carried out on 240 laying hens of Hy-Line hybrid at the age of 26 weeks. Laying hens were divided into two groups and fed with commercial diets containing 18% crude protein and 11.6 MJ metabolic energy. In the experimental group, 1% of corn was substituted with clinoptilolite preparation. During 28 research days, body mass of laying hens, laying capability, biochemical blood indicators (glucose, urea, creatinine, urates, total bilirubin, total protein, albumin, globulin, CRP, ferritin, cholesterol, triglycerides, HDL-cholesterol, LDL-cholesterol) were measured. Egg quality (n=50 per each group) was determined on day 28 of the experiment on fresh eggs, and after 14 days of keeping eggs on +4°C the following was determined: egg mass, mass of albumen and yolk, shell mass and thickness, shell firmness, height of albumen, color of yolk, pH value of albumen and yolk, HU (Haugh units) and VN (value number) of eggs. Physical traits of eggs were determined. Compared to the control, the serum of laying hens in experimental group had a significantly higher (P<0.05) level of creatinine, total bilirubin, total protein, globulins, ferritin and CRP, and lower level of triglycerides (P=0.06), measured on day 14 of the experiment. The same trend was determined on the day 28; however, the differences were not significant. There were no significant differences (P>0.05) in the egg mass, thickness and firmness of shell, pH value of albumen, HU and VN in fresh eggs. In the 14-day old eggs of experimental group, there were more favorable values determined in albumen height, HU and VN (P>0.05) and significantly lower pH in egg yolk (P<0.05). The serum changes reported in this study where a clinoptilolite preparation had been fed at 1% to layers are not adverse. Furthermore, the test substance is an activated clinoptilolite and results from this study are not directly applicable to the clinoptilolite described in the present notification.

Kaya et al. (2013) investigated the effect of a zeolite (composition not provided), an organic acid mixture and the combination of both when added to a layer diet on performance, egg quality traits and some blood parameters. The hens were fed a basal diet, a basal diet with 20 g/kg clinoptilolite, a basal diet with 2 g /kg organic acid mixture (70% propionic acid, 5% citric acid and 25% soft acid), and a basal diet with 20 g/kg zeolite and 2 g/kg organic acid mixture. Feed intake, egg production, egg weight, feed conversion ratio and body weight did not statistically differ among the groups (p>0.05). Egg quality parameters of zeolite fed animals were not affected. Dietary treatment with zeolite reduced serum albumin, glucose, ALP, AST and calcium contents compared with the control group, but treatment did not affect serum cholesterol, total protein and phosphorus levels. This study was performed using a clinoptilolite for which the purity was not given and at a higher level than 1%, therefore the results are not directly applicable to the present Notice.

Saçakli et al. (2015) determined the effect of clinoptilolite alone or in combination with phytase on the performance, carcass characteristics, intestinal histomorphology, and determined tibia ash and Ca, P levels in male broiler chickens. A total of 192 one-day-old male broiler chicks (Ross 308) were randomly assigned to 4 different treatments with 8 replicates containing 6 birds each. The control group was fed a diet without clinoptilolite and/or phytase. group 2 was supplemented with 2% clinoptilolite. Group 3 received feed supplemented 0.01 % phytase and 2% clinoptilolite, and group 4 just 0.01 % phytase.

The results showed that combined use of clinoptilolite and phytase in broiler diets decreased body weight gain at 1-14 days. Dietary treatments had no effect on body weight gain, feed intake and feed conversion ratio during the entire experimental period. Tibia ash and phosphorus level were decreased due to combined use of clinoptilolite and phytase in comparison to those fed control and clinoptilolite only diets on day 21. At 42 days of age, tibia ash, Ca, P levels and Ca/P ratio was not changed by the treatments. The authors concluded that there might be a positive effect of 2% clinoptilolite on the performance of male broilers after starting period. The purity of the clinoptilolite used in this study is not given, and thus results of the study are not directly applicable to clinoptilolite subject to this Notice. Nevertheless, the study generally supports that clinoptilolites as a group are safe when used in poultry diets at a level of 1%.

Scheideler (1993) investigated the efficacy of four different aluminosilicates (two of them clinoptilolites; all added at 1%) on aflatoxin B1 toxicity. Due to the complicated study design, it is not easy to discern results for the aluminosilicates only from those resulting from the interaction with aflatoxin B1. Body weight, feed consumption and feed conversion were not affected by any of the aluminosilicates, and calcium, phosphorus, sodium, and potassium levels in serum were not affected either. The author concludes that the type of aluminosilicate had no effect on serum calcium, phosphorus, sodium or potassium, compared with the control diet. A statistically significant reduction of chloride serum levels is reported. However, the control had 102 mEq/L, and for the four treatment groups levels of 97, 98, 100 and 101 mEq/L were reported (the author notes that the diet had not been adjusted for chloride imbalance due to addition of aluminosilicates). Bone ash was reduced by one synthetic aluminosilicate, but not by the two clinoptilolites used in the study.

Elliot and Edwards (1991) showed that adding 1.0% natural zeolite to the diet had no effect on egg weight, egg production, plasma calcium, plasma phosphorus or on the retention of calcium, phosphorus, and phytin phosphorus.

The lack of adverse effects on feeding clinoptilolite to poultry is well-supported by numerous other studies where clinoptilolite was added to diets for broilers at 1.5 to 10% (Nakaue and Koelliker, 1981; Oguz and Kurtoglu, 2000; Oguz et al., 2002; Ortatatli et al., 2005; Kavan et al., 2013; Parizadian et al., 2013; Wu et al., 2013a-d), layers at 1.5 to 4.5% (Utlu et al., 2007; Moghaddam et al., 2008; Gezen et al., 2009) and quail at 5% (Parlat et al., 1999).

6.2.4 Cats and Dogs

Roque et al. (2011) evaluated the effect of adding clinoptilolite to diets of cats at levels of 0.5%, 0.75% and 1.0% of dry matter on the digestibility and blood parameters of domestic cats. Twenty-one mongrel, adult, vaccinated and dewormed male and female cats (average age of 3 ± 0.84 yr old, average body weight 3.71 ± 0.84 kg) were housed in metabolism room and kept in individual metabolism cages. No differences ($P>0.05$) in apparent digestibility were observed between diets for dry matter, crude protein and mineral matter. The mean values for hemoglobin, indirect bilirubin and urea in blood samples of cats receiving diets containing the different additive levels were not significantly different.

Santos et al. (2013) carried out a study in which the effect of feeding clinoptilolite on the acceptability/dry matter intake of feed and apparent indigestibility coefficient (AIC) for calcium, phosphorus, magnesium, sodium, potassium, mineral excretion in dogs were measured. Fourteen adult beagle dogs (7 animals per group, average 4.5 years and body weight of 12.5 (±1.46) kg, were fed a with a standard control diet and the same diet supplemented with 1% clinoptilolite for 10 days (5 days adaption and 5 days feces collection). No effect on feed intake/acceptability was observed and there were no differences ($P>0.05$) on the production of feces (g/day) and the percentage of water in the feces. Differences were observed between the control and clinoptilolite supplemented diets for the AIC of calcium and phosphorous, but they were not significant ($P<0.02$). The AIC for magnesium in the clinoptilolite diet was less than the control ($P<0.001$). It was postulated that the reduced absorption might have occurred because the increase in the excretion of phosphorus may have led to the formation of complexes with magnesium. The AIC for potassium and sodium in both diets was similar. Similar results for palatability/digestibility and mineral excretion of diets containing 0.5%, 0.75% and 1% clinoptilolite are reported for cats and dogs (Maia et al., 2010; Santos et al., 2011).

Santos et al. (2016) also provide data on the apparent digestibility and urinary pH of adult dogs fed a commercial diet supplemented with 1% clinoptilolite. Twenty-one healthy male and female Beagles (average 4.5 yr and body weight 12.5 (±1.5 kg) and were used. No significant differences (P>0.05) were observed in the apparent digestibility coefficients of dry matter, organic matter, crude protein, gross energy, or apparent digestible energy of the control and clinoptilolite diets. Likewise, the addition of clinoptilolite to the diet had no effect on urinary pH, a finding similar to that reported for cats by Roque et al. (2011).

6.2.5 Other species

Demirel et al. (2011) conducted a study to determine the effects of dietary natural clinoptilolite (95%) on serum contents, health status and feeding performance of rats. Adult male Sprague–Dawley rats (n=24) were randomly divided into four groups with three replicates including a control group (without clinoptilolite) and three levels of natural clinoptilolite (2%, 4% and 6%) added to balanced nutritive diets. All rats were fed the above diets during 56-day experimental period. Blood samples were collected from each animal at the end of the experiment. Dietary clinoptilolite increased serum albumin, triglyceride and VLDL levels (P<0.05). However, the differences among treatment groups were not significant for serum minerals (Ca, P, Mg, K, Na, Cl, Fe), urea, Fe binding, LDL, alkaline phosphatase, glucose, uric acid, total Fe, total protein, globulin, cholesterol, HDL cholesterol, creatinine; metabolizable energy and crude protein consumption for 1g live weight gain of rats (P>0.05). The authors concluded that dietary use of clinoptilolite at 2, 4 and 6% did not cause any adverse clinical and biochemical effects on the health status of rats.

Demirel et al. (2015) investigated the effects of dietary natural zeolite (clinoptilolite) on performance, skin and liver histology in male rats. Animals were divided into four groups with three replicates, including a control group and groups with one of three doses of clinoptilolite (2%, 4%, and 6%) in their diets. All the rats were fed these concentrates throughout the experimental period of 56 days. There were no significant differences in the primary and secondary hair follicle numbers among groups, but the change in diameters of each follicle were found to be significant. The primary and secondary follicle numbers and diameters ranged from lowest to highest as follows: 2.00-2.33, 4.50-7.17, 11.53-20.42 and 57.63-102.12 μm, respectively. The differences occurred between the control group and group IV (containing 6% zeolite). In addition, the skin and liver histology results showed that there were no differences among the groups. Though the clinoptilolite used in this study was of different origin, it is comparable and supports in general terms the safety of the clinoptilolite described in this notification.

Demirel et al. (2018) compared the effects of dietary clinoptilolite on nutrient digestibility and relative organ weights in rats. In their study, 24 adult male Sprague-Dawley rats were divided into 4 groups with 3 replicates, according to a randomized split plots design. In addition to a control group (0% clinoptilolite), 3 levels of clinoptilolite (2%, 4%, and 6%) were used in the diets, and the rats were fed these diets for 56 days. Statistically significant differences were found among the groups for the digestibility rates of nutrients (P<0.05), except for crude fiber and acid detergent fiber. The addition of clinoptilolite in rats' feeds reduced the digestibility of crude fiber, crude ash, neutral detergent fiber and acid detergent fiber, but increased the digestibility of other nutrients. Slaughter live, organs (liver, kidney, heart, and stomach) and relative organ weights were not affected significantly by the clinoptilolite contents. The authors concluded that clinoptilolites can be used in animal feed safely at levels of up to 6%. Though the clinoptilolite used in this study was of different origin, it is comparable and supports in general terms the safety of the clinoptilolite described in this notification.

As stated previously, In view of the numerous sources of clinoptilolite and their wide range of applications, it is important to evaluate whether those studies presenting evidence for hazards when clinoptilolite is added to animal diets are comparable to the substance and conditions of use of the clinoptilolite described in this Notice. Again, with reference to a study considered by EFSA (2013), Martin-Kleiner et al. (2001) supplied young adult mice with food containing 12.5%, 25% or 50%

clinoptilolite powder. Control animals received the same food without the clinoptilolite. Clinoptilolite ingestion was well tolerated, as judged by comparable body masses of treated and control animals. A 20% increase of the potassium level was detected in mice receiving the zeolite-rich diet, without other changes in serum chemistry. No other parameter showed a deviation despite the high feed inclusion level. Data obtained at diets inclusion levels of 12.5 % or higher serve for hazard identification, whereas as such moderate changes have not been observed at 2% or lower. This finding, deemed by EFSA (2013) to present a possible hazard, is not relevant for the risk assessment of the proposed conditions of use of the clinoptilolite in this Notice.

Additionally, EFSA (2013) refers to two studies performed by Gerasev et al. (2003a,b) who described reduced potassium absorption and increased renal excretion in rats. In these studies, a clinoptilolite (from Siberia) was added at 5% to the diet and fed to male rats for a short period of seven days. This study's design is inadequate to study long term administration of clinoptilolite to rodents, moreover the level it was added to the diet is significantly above 1%.

6.3 Feeding studies in target animals with Zeocem clinoptilolite

The first three sub-sections review studies in target animals that were carried out using clinoptilolite sourced from the Zeocem mine in Nižný Hrabovec, Slovak Republic; the fourth sub-section compares clinoptilolite used in these studies with clinoptilolites used in studies discussed in section 6.2

6.3.1 Ruminants

Cattle

The effects of supplementing rations with 2 to 7.5% clinoptilolite were studied in beef cattle and dairy cows (Vrzgula et al., 1985; reviewed by Bartko et al., 1995). Long-term supplementation revealed no significant differences in weight gain, feed efficiency, or milk yield between supplemented and control animals. Similar results were reported by the same authors for lambs supplemented across this range.

Vrzgula et al. (1988) carried out an experiment to determine the effect of adding clinoptilolite to the colostrum given to calves in the control/treatment of diarrhea. A total of 43 calves (23 controls and 20 treated) from Slovak spotted and Black spotted breeds were kept on two farms. The calves individually received 2 L of maternal colostrum within the first 3 hours of life and 3x daily for 15 days. The colostrum for the treated calves was enriched with 1% clinoptilolite. General health and incidence of diarrhea were monitored daily. Blood was taken immediately after birth and at 24 hr, and on days 5, 10 and 15 after parturition. Total protein and immunoglobulin levels, and serum concentrations of Ca, Cu, Fe and Zn were measured.

The results showed that clinoptilolite added to the colostrum given to the calves produced an increase in the concentration of total immunoglobulin in the blood serum of the treated groups, but the increase was significant only on one farm on days 5 and 10. An increase in the serum levels of total proteins in treated calves compared with the controls was observed in each collection beginning 24 hours after birth. This difference was statistically significant ($P<0.01$) on day 10. Calcium concentration increased ($P<0.05$) in the treated groups on the day 10 and it persisted until day 15 on Farm 2. Different results were noted in both farms in the analysis of serum P which was higher in the treated groups in each collection than in the control groups on Farm 2; the situation was similar in the first collections in Farm 1, but on day 15 there was a statistically significant ($P< 0.01$) decrease in the mean concentration of the serum P in the treated groups compared with the controls. Serum Fe level of treated calves increased markedly 24 hours after parturition on both farms. There were significant differences in favor of the treated groups in the level of copper, showing the highest concentrations on days 10 and 15. At the same time, there was an increase in the concentration of the total proteins which are the biological carriers of trace elements, forming together protein metal enzymatic complexes. There were no significant differences in serum zinc on either farm due to clinoptilolite. Throughout the course of the experiment the occurrence of the diarrhea syndrome was recorded on both farms in the treated and control groups; in the latter diarrhea developed in 13 calves. In five cases it was complicated by respiratory syndrome ending in death. In the treated groups the course of the diarrhea was moderate and lasted only 2 days. These results confirm an earlier trial (Vrzgula, 1985) where feeding clinoptilolite (1% of diet) twice daily to calves for 10 to 14 days reduced the incidence of diarrhea. Adding clinoptilolite (2% of diet twice daily) was found to have a positive effect on animals already affected with the syndrome.

Sheep

Bartko et al. (1983a) studied the effect of clinoptilolite on the health condition of 10 sheep of the Merino breed; a control group (5 animals) fed a basic diet and experimental group (5 animals) given the same diet with 1.5% clinoptilolite for 3 months. No significant differences were observed in the health condition and general behavior of the control animals and those fed clinoptilolite. Differences observed in content of volatile fatty acids in rumen contents, blood chemistry, micro/macro elements, transaminase activity and acid-base homeostasis were also not significant.

6.3.2 Swine

Vrzgula et al. (1984) carried out a feeding trial with clinoptilolite in a large-scale fattening facility with 450 pigs in both the control and treatment groups. The pigs (crossbreed Slovak white pedigree x Landrace) weighed an average 22 kg in the control and 20 kg in the experimental group. Animals were fed a starter ration until reaching about 45 kg and then a finisher ration. Clinoptilolite was added to both rations of the experimental group at a level of 3%. The feeding mixtures were given in troughs in granulated form ad libitum. Animals were weighed on days 70 and 166. General observations of behavior and sensory evaluation of feces was carried out during the 167 days fattening period. At the beginning of fattening the-average pig weight in the experimental group was about 2 kg lower than in the control group. However, after 70 days, it was 4.14 kg higher in the experimental group than in the control. At the end of the fattening the average weight of the experimental group was 3.96 kg lower than the control. The average daily weight gain in the experimental group over 70 d, was 13.5% higher than in the control, however during the remaining 96 days (until the end of fattening), the average daily weight gain was 5.8% lower in the experimental group than in the control. Overall, the average daily weight gain of the entire period of fattening was only about 1% higher in the experimental group. The mortality rate of the pigs receiving clinoptilolite was 2.9% lower than the control animals. The feces of pigs receiving clinoptilolite were denser, lighter and smelled less than the feces of control animals.

Vrzgula et al. (1982, 1984) and Bartko et al. (1983b, 1995) also report data from large-scale commercial studies in which experimental groups of 100 to 600 swine were fed diets supplemented with 5% clinoptilolite for 30 to 45 days. No detrimental effects on animal performance and health were reported for any of the studies.

6.3.3 Poultry

Herzig et al. (2008) conducted a study to find out the impact of a long-term application of clinoptilolite on the chemical composition of long bones (femur, tibiotarsus) and eggshell of layers. A total of 120 birds of the hybrid breed Bovans Goldline were divided into a control group fed a standard feed mixture and an experimental group in which the diet was supplemented with 1% Zeocem clinoptilolite (ZeoFeed®). The layers were raised in a three-floor cage structure with automatic watering and manual feeding in an environment with a regulated lighting and thermal schedule. The actual experimental period started in the twenty-second week of age of the layers and ended in the sixty-eighth week. Layers receiving the clinoptilolite had higher average egg weights ($P<0.05$) and the consumption of feed mixture per one egg was 4 g lower compared to the control birds. The eggshell of layers receiving clinoptilolite had a higher ($P<0.01$) content of crude protein, calcium and magnesium than the controls. The values of the same indicators were higher in the femur and tibiotarsus ($P<0.05$ and $P<0.01$, respectively) of layers fed clinoptilolite. The health condition of the layers was monitored daily and no clinical symptoms of a disease were registered. It was reported that the long-term application of clinoptilolite favorably influenced the lodgment of Ca, P, Mg and crude protein in the eggshell, as well as in both long bones, increased egg production and reduced the consumption of feed mixture per/egg, while maintaining health of the layers.

Suchý et al. (2006) carried out trials to determine the effect of supplementing broiler diets with Zeocem clinoptilolite (ZeoFeed®) at levels of 1% and 2% on growth performance. The average liveweight of supplemented birds was higher ($P<0.01$) than for the control group at 40 days of age. There were no differences in water consumption between groups at either level of supplementation. Chickens in all groups showed very good health and a very low rate of mortality.

Other studies where Zeocem clinoptilolite (ZeoFeed®) was fed to layers include Straková et al. (2008) who examined at a feeding level of 1% selected production parameters, and hematological and biochemical parameters of hepatic and renal function. In the course of the experimental period from week 22 to week 68, the layers receiving clinoptilolite had a 1.7% higher laying intensity as compared to control layers (i.e. the number of laid eggs in experimental layers increased by 5.6 eggs per layer). The mean weight of all laid eggs was 66.3 (±6.25) g in the control group and 65.6 (±5.44) g in the experimental group. Consumption of feeding mixture per egg laid was 141.7 g in the control group and 137.6 g in the experimental group. The consumption of feeding mixture in the experimental group was 4.1 g lower than that in the control group. The mean values of parameters monitored in blood plasma, such as uric acid, cholesterol, glucose, lactose, calcium, phosphorus, ALP and LDH, in both groups of layers ranged within reference intervals, with no significant differences being detected between both groups. Statistically significant differences between both groups were found in total plasma protein ($P\leq0.01$), triacylglycerol levels ($P\leq0.05$), and magnesium ($P\leq0.01$), which were elevated in the control group, and in AST ($P\leq0.05$) whose level in the control group was significantly lower than that in the experimental group. The results of hematological tests performed with layers' blood revealed statistically significant changes in parameters such as the erythrocyte count ($P \leq 0.01$), hemoglobin level ($P\leq0.01$), and MCHC ($P\leq0.05$), which were elevated in the experimental group, and in the leukocyte count ($P\leq0.05$), which was lower in the experimental group, as compared with the control. However, serum value changes found varied within physiological ranges and do not represent adverse effects raising safety concerns. They may very well reflect differences in feed intake and egg production.

Machacek et al. (2010) conducted a study to assess the effects of two levels of clinoptilolite administered in feed (2% and 4%) on some selected performance indicators, metabolic utilization of basic nutrients and the health status of laying hens. The selected 24 Bovans Goldline hybrid laying hens were divided into three equal groups, the two experimental groups and one control group. The laying hens were housed individually in cages with an automatic supply of drinking water, manual feeding, in a setting with controlled light and temperature regimens. Hens from individual groups were all fed a complete feed mix of the same composition and the only difference was in clinoptilolite supplementation: feed mixes for supplemented groups that contained 2% and 4% of clinoptilolite (commercial additive ZeoFeed) respectively, replacing the same amounts of wheat. The hens received feed mixes and drinking water ad libitum. During this 28-day experiment, feed consumption and the number and weight of eggs laid were monitored individually for each hen. At the end of the experiment, the balance test using the indicator method (Cr_2O_3) was performed in all eight hens in each of the groups. The results of balance tests were then used to calculate the metabolic utilization of selected nutrients (nitrogen, fat, ash, nitrogen-free extracts, starch, gross energy, Ca, P). After the balance tests, blood samples for hematological and biochemical examinations were collected via puncture of the vena basilica. The addition of 2% clinoptilolite to feed mix resulted in a highly significant ($P\leq0.01$) increase in mean egg weight to 64.69 g, but the addition of 4% clinoptilolite resulted in a highly significant ($P\leq0.01$) decrease in mean egg weight to 62.20 g compared to the control (63.73 g). Moreover, daily feed mix consumption with 2% clinoptilolite decreased to 114 g per one laying hen/day compared to the controls (118 g per one laying hen/day). In the 4% clinoptilolite), daily consumption of the feed mixture increased compared with the controls to 124 g. The 2% clinoptilolite supplementation feed slightly increased metabolic utilization of fat, nitrogen-free extracts, starch and gross energy compared to control group. Results of hematological test of the hens' blood showed significant changes in hemoglobin, whose values in control and 2%-groups were significantly higher ($P\leq0.05$) than in the 4%-group. Differences in the values of the biochemical indicators monitored (total protein, glucose, cholesterol, triacylglycerols, calcium, phosphorus, magnesium and the AST enzyme) between the control and both experimental groups were not significant and remained within the range of reference values.

6.3.4 Applicability of studies with other clinoptilolites (6.2.) to Zeocem clinoptilolite

The studies reviewed in Sections 6.2 and 6.3 demonstrate that clinoptilolite can be added to the diets of ruminants, swine and poultry under normal production conditions at levels of 1% or more without eliciting detrimental effects on animal health or overall performance. The read-across between other clinoptilolites studied in target animals and clinoptilolite described in this GRAS notice requires a comparison of their composition.

A comparison between the composition of the Zeocem clinoptilolite which is the subject of this notice and the composition of the clinoptilolite used in some of the feeding studies cited in Section 6.2 is given in Table 6. 1. The data demonstrate that the purity and mineral composition of the oxides of the Zeocem clinoptilolite is very similar to that of the clinoptilolite used in the reported animal feeding trials, and for which no safety concerns were indicated for animal performance or health.

Table 6. 1	**Similarities between Zeocem clinoptilolite and clinoptilolite used in feeding studies**						
Reference	Zeocem[a]	Katsoulos et al. (2005a,b,c)	Karatzia et al. (2011)	Pourliotis et al. (2012)	Papaioannou et al. (2002)	Sardi et al. (2002)	Herzig et al. (2008)[b]
Animals	Ruminants, swine, poultry and pets	Dairy cows	Dairy cows	Calves	Swine	Swine	Poultry
Parameter							
Purity (%)	86 - 92	92	92	92	88[c]	≥85	>80
Mineral composition (%)[d]							
SiO_2	65.0 - 71.3	68.9	69.9	68.9	68.26	68.20	62
Al_2O_3	11.5 - 13.1	11.3	11.3	11.3	13.30	12.3	14
CaO	2.7 - 5.2	3.0	3.0	3.0	4.34	4.34	5.5
Na_2O	0.2 - 1.3	0.75	0.75	0.75	0.26	0.26	NG
Fe_2O_3	0.7 - 2.1	0.11	0.11	0.11	0.08	0.08	2.3
K_2O	2.2 - 3.4	2.23	2.23	2.23	0.94	0.94	NG
MgO	0.6 - 1.2	0.6	0.6	0.6	1.05	1.05	NG
Si:Al (ratio)	4.8 - 5.4	6.1	6.1	NG	5.1	5.5	4.4
Particle size (mm)	0.01 - 2.5	0.80	NG	<0.80	NG	0 - 1	0.2 - 0.5

[a] Clinoptilolite sourced from Zeocem's mine in Nižný Hrabovec, Slovak Republic (Appendices 2.1 and 2.2).
[b] Clinoptilolite used in the study was a Zeocem clinoptilolite ZeoFeed® product.
[c] Estimated amount; calculation based on data provided in study.
[d] Mineral composition of reported oxides.
NG - Not given.

It may be questioned whether the clinoptilolites from other sources used in the studies discussed in Section 6.2 are also comparable to the substance that is the subject of the present notice with respect to impurities and contaminants.

For heavy metals, arsenic, and dioxins/dioxin like PCBs maximum limits for complete feed were established in Europe between 1974 and 2007; in addition, for specific types of feeds such as mineral feed or feed ingredients such as feed additives derived from mineral deposits (among them clinoptilolite of sedimentary origin) separate levels were adopted. All these levels reflected and, as they have been reviewed regularly, continue to reflect the state-of-the-art toxicology and good manufacturing practice (which included reported and observed ranges of contamination).

The maximum limits were applied historically in Europe (many as early as 1974, most of them before 2003) and apply today for complete feed, complimentary feed (mineral feed), and for clinoptilolite. [3] As comparison maximum tolerable levels for heavy metals and arsenic based on animal health as adopted by NRC are given in Table 6.2.

Table 6. 2 Maximum levels for contaminants in feed (European Union)

Matrix	Lead	Cadmium	Mercury	Arsenic	Dioxins	Dioxin and dioxin-like PCBs
	mg/kg				ng WHO - TEQ/kg	
Complete feed	5	1	0.1	2	0.75	1.5
Complementary feed	10	0.5	0.2	4	0.75	1.5
Mineral feed	15	5	0.2	12	-	-
Clinoptilolite	30	2	[0.1]	[2]	0.75	1.5
Maximum Tolerable Levels (NRC 2005)	10 – 100	10	0.2	30	-	-

Note: levels in [ ] are not legally mandatory, but respected and applied by the feed chain.

The safe/unsafe intake of a contaminant for an animal results from all dietary sources, including water for drinking. As long as a single ingredient's content of a contaminant is not higher than what is tolerable for the complete feed or ratio, the resulting net intake by the animal would continue to be safe. Should a higher content be permitted in an ingredient, a dilution in the complete feed to levels below the tolerance for complete feed would assure that intake by the animals is still safe.

Study protocols for animal studies are prepared under applicable regulatory framework including rules for animal welfare and animal feed; they reflect also current feeding practice in the country where the study is run.

It is expected that levels of contaminants in the complete feeds used in the studies discussed in Section 6.2 are, when performed in Europe, meeting the legal requirements laid down in European Union feed law. This applies specifically to the studies run in Europe which are those reported by Katsoulos et al. (2005a,b,c), Karatzia et al. (2011), Pourliotis et al. (2012), Papaioannou et al. (2002), Sardi et al. (2002), and Herzig et al. (2008). The clinoptilolites used in European studies are therefore similar to the clinoptilolite subject to this notification with respect to their likely contamination with heavy metals, arsenic and dioxins, as they all were products used under applicable European feed law which includes levels for these contaminants. Such studies are done to develop data to be used in the market place which makes only sense if the product comply with regulations and can therefore be marketed.

However, also studies with clinoptilolite run outside of the European Union under a different jurisdiction would support the conclusions on the safety of the clinoptilolite. If results from such studies indicate safety of the applied clinoptilolites at the proposed use level of 1% or higher, the study material used was at least contaminated at levels that did not results in toxicity. The notifier is not aware of studies with reported toxicity of clinoptilolite due to presence of contaminants, the materials used in the cited studies are considered to be also sufficiently similar with respect to their spectrum and level of contaminants.

In summary, other clinoptilolites used in those studies discussed in Section 6.2 are similar to clinoptilolite of this notice with respect to their major constituents and their content in heavy metals, arsenic, and dioxins/dioxin like PCBs.

[3] DIRECTIVE 2002/32/EC OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 7 May 2002 on undesirable substances in animal feed (as amended subsequently).

6.4 Animal Safety: toxicological studies

6.4.1 Bioavailability and absorption

Demirel et al. (2011) conducted a study to determine the effects of dietary natural clinoptilolite (95%) on serum contents, health status and feeding performance of rats. Adult male Sprague–Dawley rats (n=24) were randomly divided into four groups with three replicates including a control group (without clinoptilolite) and three levels of natural clinoptilolite (2%, 4% and 6%) added to balanced nutritive diets. All rats were fed the above diets during 56-d experimental period. Blood samples were collected from each animal at the end of the experiment. Dietary clinoptilolite increased in serum albumin, triglyceride and VLDL levels ($P<0.05$). However, the differences among treatment groups were not significant for serum minerals (Ca, P, Mg, K, Na, Cl, Fe), urea, Fe binding, LDL, alkaline phosphatase, glucose, uric acid, total Fe, total protein, globulin, cholesterol, HDL cholesterol, creatinine; metabolizable energy and crude protein consumption for 1g live weight gain of rats ($P>0.05$). The results showed that the supplementation of clinoptilolite did not have positive effect on serum concentrations of the investigated parameters apart from albumin, triglyceride and VLDL, but they had no negative effect on the health status of animals.

Studies of the bioavailability of clinoptilolite minerals in porcine models have been conducted by numerous investigators. Pond and Yen (1983a) reported that the concentrations of liver and kidney ash, Cd, Zn, Fe, Mn, Cu, Ca, Mg, P, K, and N were unchanged in growing pigs fed a diet containing 3% clinoptilolite, thereby indicating that clinoptilolite is considered to be stable at physiological pH and at the acidic pH (<2) of the stomach. Shurson et al. (1984) fed pigs diets containing up to 7.5% clinoptilolite. Analysis of feces and urine samples for mineral retention indicated a reduction in levels of P, but no changes in the levels of Ca, Mg, Na, K, Fe and Zn. The authors stated that this indicated the stability of clinoptilolite in the acid environment of the stomach. In another study Pond et al. (1989) reported that feeding growing pigs 2% clinoptilolite for 12 weeks had no consequence on the concentrations of P, Fe, Zn, or Al in either kidney or liver tissue. The authors stated that the failure of liver or kidney Al to be affected by supplemental clinoptilolite in the diet suggests that clinoptilolite is stable in the milieu of the digestive tract contents and therefore, the absorption of Al or Si contained in the crystalline matrix of the molecule is nil or low.

Papaioannou et al. (2002) reported that supplementing sow diets with 2% clinoptilolite had no significant effect on vitamins (A and E), and minerals (K, Na, P, Ca, Mg, Cu, and Zn) uptake and/or distribution. Fokas et al. (2004) reported that supplementing the diets of castrated male pigs with 2% clinoptilolite did not lead to a significant increase of Pb concentration in blood or edible tissues, except for liver ($P <0.001$). However, the Pb retention coefficient between diets was not significant. The retention coefficient of Pb in the clinoptilolite used was practically negligible (0.009). The amount of Pb contained in the supplemented diet (2.1 ppm) was below the allowable EC limit of 5 ppm for complete feeds (EC, 2002) and well below the level of 30 ppm given by NRC (2005) for mineral feeds.

Alexopoulos et al. (2007) studied the effect of long-term use of clinoptilolite on certain biochemical and hematological parameters in piglets. Forty-eight piglets (24 females and 24 males), equally allocated in two experimental groups were fed a control diet or the control diet with 2% clinoptilolite continuously from weaning to slaughter. All pigs were individually weighed, and blood samples taken upon commencement of the study (25 days of age) and subsequently at the end of the weaning, growing and fattening stages (70, 112 and 161 days of age), respectively. The diet had no effect on serum K, Na, Ca, P, total protein, albumin and total bilirubin concentrations throughout the study ($P<0.05$). Likewise, no significant alteration was noticed concerning the hematocrit, leucocyte count and hemoglobin concentration ($P<0.05$).

Dwyer et al. (1997) conducted a study in which day-old broiler chicks were fed a diet containing 1% clinoptilolite until 3 weeks of age. No significant differences were observed in hematology, serum biochemical values and enzyme activities. Similar results for blood serum biochemical parameter are reported by Miles and Henry (2007) who fed broilers diets containing 2% hydrated sodium calcium aluminosilicate for 21 days and Safameher et al. (2008) and Eleroglu et al. (2011) who fed broiler chicks diets containing 2% and 5% clinoptilolite, respectively for 42 days.

In summary, available research indicates that clinoptilolite is not soluble and is resistant to the physiological and chemical conditions in the digestive systems of monogastric and ruminant animals; it passes through the gastro-intestinal tract essentially unabsorbed and is excreted with the feces. No studies could be found in the scientific literature demonstrating that clinoptilolite is degraded or absorbed after being consumed, and thereby posing a health risk for animals. As previously noted, it is already authorized for use as an anticaking agent in Europe (EC, 1999; EU, 2011; EFSA, 2013). This conclusion is supported by further data discussed in the following Section 6.4.2.

6.4.2 Stability of clinoptilolite in simulated gastric and other relevant fluids

Clinoptilolite is chemically highly stable under acidic conditions due the abundance of high energy silicon-oxygen bonding throughout the tectosilicate structure (Cotton, 2008).

Available evidence indicates that clinoptilolite resists degradation by gastric and intestinal juices. In studies using simulated body fluids, the crystal structure of clinoptilolite is reported as remaining stable (Rivera et al., 1998; Ceyhan et al., 2007), even up to 40 days (Martin-Kleiner et al., 2001).

Alver (2013) investigated HCl-treatment of clinoptilolite and demonstrated that under different pH conditions (from 0.2 to 2 M HCl at 70 °C) the amount of iron in the material stays constant with respect to the original material. Moreover, it could be proved that the treatment of clinoptilolite with hot acid did not cause any considerable effect/damage on the crystal lattice of the mineral structure.

Waluyo et al. (2018) investigated a modified natural zeolite containing clinoptilolite, heulandite (structurally equivalent to clinoptilolite) and mordenite. They reported that the treatment of the material with ammonium nitrate followed by acid treatment over several hours did neither significantly alter the structure of the included clinoptilolite phases nor the content of Fe.

Akkoca et al. (2013) treated clinoptilolite with acids (hydrochloric, phosphoric and bromic) and alkalis (potassium hydroxide and sodium hydroxide). X-ray diffraction analysis indicated that the structure of clinoptilolite was not altered by either of the treatments.

Ates and Hardacre (2012) reported that even if the ion composition of clinoptilolite is altered by ion exchange processes and a following heating step, treatment with 1M HCl for 30 minutes afterwards did reduce the Al concentration of clinoptilolite by less than 10%. Fe concentration was not altered significantly. The authors concluded that upon ion-exchange (...) Fe^{3+} and Mg^{2+} can only be partially removed due to its stronger binding in clinoptilolite (...) and the extent of dealumination towards acid leaching is low.

Polatoglu (2005) incubated clinoptilolite with simulated gastric juice for 2 hours at 37°C, using a finely ground clinoptilolite with a particle size of 24 µm. Incubation revealed a leaching rate of <3% for Al and <4% for Fe.

Beltcheva (2015) performed some experiments with rats, using a modified form of clinoptilolite, adding it to a 10% alkaline salt solution and processing the mixture with addition of 25% water for 4h in a ball crusher. During this process, < 10% of the Al became available, whereas Fe was leached by 55%.

Li et al. (2008) estimated the stability of clinoptilolite by measuring the leaching of Al, Si, K and Na ions after acid treatment (pH ranging from 1.0 to 5.0) over periods of 24 hours to 6 days. They reported that clinoptilolite was chemically stable at low pH environments, as only 11% of Al was leached even after incubation at pH 1 for 144h. At pHs of 3, 4 and 5 the leaching of Al decreased drastically.

In a study performed by Troszkiy (2017) it was shown that even with boiling 5% HCl only 14% of Al and 0% Fe could be leached from clinoptilolite. Even more rigid conditions were used by Kadirbekov et al. (2017) who treated clinoptilolite with 1.75N HCl at 96-98°C for 6 hours. This led to a leaching of only 1/3 of the Al from the clinoptilolite lattice and 75% of Fe.

Rivera et al. (2013) performed some studies with clinoptilolite and different HCL solutions with a solid liquid ration of 1g/250ml for 1 hour at room temperature. Al content was reduced with a physiological

HCl concentration of 0.1N from 5.9% (control) to 5.3 %. The author states that the structural stability of clinoptilolite was not significantly affected by acid treatments.

Kavak and Ülkü (2012) also investigated the structural stability of clinoptilolite in simulated digestion conditions and their interactions with digestive media and human colon adenocarcinoma cells. Analyses (XRD, fourier transform infrared spectroscopy, scanning electron microscopy and inductively coupled plasma atomic emission spectroscopy) of the clinoptilolite samples showed that the substance preserved its structural stability during in vitro digestion.

Leung et al. (2007) conducted laboratory tests to determine the stability of a zeolite (containing 90% clinoptilolite, high in Al, Fe, Pb) under acidic conditions (pH 1.5) similar to those found in the stomachs of swine. It was reported that after 25 hours the amounts of the heavy metals released were well below the toxic levels as recommended by NRC (2005).

6.4.3 Reduced bioavailability of Al in presence of Si

The bioavailability of aluminum from food and water sources is low, estimated to range between 0.1% and 0.4% (WHO, 1997; Krewski et al., 2007). An important limitation to the bioavailability of aluminium is the interaction with aqueous aluminium species and dissolved silica; silicic acid $Si(OH)_4$ generating hydroxyaluminosilicates (Birchall, 1992).

Interestingly, many studies showed that this figure is further decreased upon co-supplementation with silicon (Domingo et al., 2011; Birchall, 1992,1995; Edwardson et al., 1993). Based on epidemiologic data, several authors performed human studies on volunteers to try and establish a relationship between the presence of soluble silicon in drinking water and the bioavailability of aluminum upon ingestion. Edwardson et al. (1993), using labelled Al on 5 volunteers, showed a reduction of 85% of peak plasma aluminum concentration upon administration of soluble Si in drinking water. In a similar experiment, Bellia et al. (1996) measured the urinary Al excretion in volunteers given beer with a high Si content. The authors calculated a reduction of 85% of the bioavailability of Al (estimated at 0.09% after 1 hr) in the presence of silicic acid compared to controls. Davenward et al. (2013) and Jones et al. (2017) showed increased Al urinary excretion in volunteers who drank a silicon-rich mineral water for 12 weeks. Interestingly, iron and copper urinary excretion remained unaffected by the treatment.

The data are supported by a study by Kraljevic-Pavelic et al. (2017) performed in rats. Rats were gavaged daily with a solution of aluminum chloride, followed after 1 hour by a new gavage solution containing colloidal silica or different types of clinoptilolites. Control groups received only aluminum chloride, only colloidal silica or only zeolite. The authors concluded that the results clearly showed increased Al concentrations in the plasma and target organs, namely liver and bones of Al-intoxicated animals upon supplementation with aluminum chloride that were markedly decreased in Al-intoxicated animals supplemented with clinoptilolite materials. The same effect was observed in Al-intoxicated animals supplemented with colloidal silica.

6.4.4 Carcinogenicity

Studies lasting up to one year with Sprague-Dawley rats have demonstrated that intratracheal injection of 10 to 15 mg of clinoptilolite dust (50% clinoptilolite content, particle size of <2 μm) did not lead to significant damage of macrophage cell membranes. Mild fibrosis was detected, but no significant rise in tumor incidence. None of the doses applied caused heavy fibrosis or mesothelioma (Adamis et al., 2000). Likewise, no significant dose-related increase was found in the incidence of tumors in any organ or tissue of Wistar rats given intratracheal doses of clinoptilolite (0, 30, or 60 mg/animal) to groups of 60 or 50 male and 50 female rats on one occasion (Tátrai and Ungváry, 1993; WHO, 1997). In vitro cell culture studies have shown that clinoptilolite, unlike other zeolites, caused no alveolar macrophage cytotoxicity due to the shape of its dust particles (Dong et al., 2006). At present, clinoptilolite is placed in Group 3, being not classifiable as to its carcinogenicity in humans due to inadequate evidence in humans and experimental animals (WHO, 1997; Elmore, 2003). Thomas and Ballantyne (1992) who carried out an extensive review of toxicity of zeolites, including clinoptilolite, reported that mice did not illicit peritoneal tumors when exposed to fibrous clinoptilolite.

6.4.5 Toxicity

Pavelić et al. (2001) conducted a series of studies in mice using 4-months-old male and female CBA/HZgr mice administered diets containing 0 or 25% clinoptilolite for a period of 1, 3, or 6 months. During the course of study, the animals were monitored daily for clinical signs, changes in behavior, and survival, changes in body weight were monitored weekly, and food and water consumption measured at days 14 and 28 when the mice were housed for 24 hours in metabolic cages (5 mice per cage). Changes in hematological and serum clinical chemistry parameters (erythrocytes, leukocytes, platelets, hematocrit, hemoglobin, glucose, ALP, AST, ALT, bilirubin, inorganic phosphorus, and calcium) were obtained at t = 1, 3, and 6 months and 24 hours urinalyses (glucose, proteins, urobilinogen, bilirubin, nitrites, erythrocytes, leukocytes, pH and specific gravity) obtained monthly. Histopathological analyses of liver, spleen, kidney, brain, lung, testes, ovary, duodenum, eye, stomach, large and small intestine, muscles, myocardium, pancreas, thymus, auxillary lymph node were carried out on all animals at the end of the 1, 3, or 6 months' timepoints. The investigators reported that no changes were observed that could be considered a toxic effect of the treatment. The results of this feeding study with clinoptilolite suggest a NOAEL of 25% in the diet (about 37,500 mg/kg body weight/day), the highest dose tested.

Pavelić et al. (2001) also conducted a study using Wistar rats administered diets containing 0, 25, or 50% clinoptilolite for a period of 1, 3, or 12 months. The protocol used for these studies was similar to that presented for mice as described above. During the course of the study rats were monitored daily for clinical changes in behavior and incidence of mortality, body weight and water consumption were measured every 4 days and feed consumption measured daily. Hematological and serum clinical chemistry parameters (as per mice) were measured monthly. Histopathological examinations of liver, spleen, lung, kidney, testes ovary and brain, were performed on experimental and control rats euthanized after 1, 6, and 12 months of feeding. The investigators reported that no changes were observed that could be considered a toxic effect of the treatment. The results of this feeding study in rats with clinoptilolite suggest a NOAEL of 50% in the diet (about 25,000 mg/kg body weight/day), the highest dose tested.

6.4.6 Reproductive and developmental toxicology

In a multi-generation study Pond and Yen (1983b) investigated the effects of long-term ingestion of clinoptilolite on growth and reproduction in Sprague-Dawley rats. Female rats were administered a basal diet or a diet containing 5% clinoptilolite at weaning through week 13, at which point the females were mated and the dams continued on the same feeding regimen. Parameters such as body weight after parturition and at 1, 2, and 3 weeks postpartum, number of pups born per litter, live birth weight and average total litter birth weight were recorded. At 1, 2, and 3 weeks postnatal, the number of live pups and total litter weight were recorded. The pups were weaned at 3 weeks and the dams were euthanized. Blood samples were collected from the dams for determination of hemoglobin, hematocrit and plasma urea nitrogen. Fresh and dry weights of livers and kidneys were recorded. Pups (2/sex) from each litter were randomly selected at weaning and placed in a single cage. Individual body weights were recorded weekly for 4 weeks, after which males were euthanized, blood collected for hematocrit and liver, kidneys and testes removed and weighed. The females were mated with young adult fertile males. Pregnant females continued on their respective diets during gestation and were weighed immediately after parturition and 1 day postpartum. Numbers of total and live pups per litter were recorded, and total weight of each litter at birth and at one day of age was determined. Dams were euthanized in the same manner as the first generation, blood hemoglobin and hematocrit were determined, and liver and kidneys weighed. The authors reported that ingestion of clinoptilolite over 2 generations had no adverse effects on growth or reproduction in the animals. There was no evidence of toxicity of feeding clinoptilolite (5% in diet) during the growing period of the dams or throughout pregnancy and lactation. Reproduction was not affected by clinoptilolite consumption and no teratogenic effects or adverse effects on development or growth of the offspring were observed.

Pavelić et al. (2001) investigated the reproductive toxicity of clinoptilolite in mice. In these experiments, clinoptilolite was mixed with standard feed at a ratio of 25:75 and was administered to

10 male and 10 female CBA/HZgr mice for 50 days, starting at least 14 days prior to mating. The treatment continued during the pre-pregnancy and pregnancy period (one cycle) and until weaning. The same pair of animals was fed clinoptilolite diets and monitored during four consecutive cycles (approximately 4 to 5 months). The same schedule was applied for control animals. The parental generation was monitored for duration of the cycle period (pre-pregnancy and pregnancy period), fertility (presence or absence of litter in a particular cycle), delivery incidence, mortality, and histological changes of the ovaries, after the fourth cycle. The number of total and viable pups born, as well as gain in pup body weight and mortality until weaning also was recorded. The pre-pregnancy period was shorter in the clinoptilolite-treated mice. The number of pups per litter was increased in clinoptilolite-treated mice. The authors postulated that probably for this reason the gain in body weight until weaning was decreased and a higher rate of mortality between days 8 and 21 of the neonatal period was observed. However, there were no differences between control and treated animals that would suggest reproductive toxicity attributable to the clinoptilolite administration.

Kyriakis et al. (2002) conducted a study with 240 crossbred gilts and sows divided equally into two groups, and 2% clinoptilolite incorporated into the feed of one group. The sows and gilts of each group were offered the experimental diets from weaning (or from age of 6 months for the gilts), during service, pregnancy and lactation and up to the date of service of the next reproductive cycle. All sows and gilts and their litters were monitored daily. No adverse or side effects were noted in the sows that were on the clinoptilolite-enriched diet. They showed normal estrus behavior during the breeding period and gave rise to a slightly better farrowing rate (92.5%) compared to the control group (85.8%). No teratogenic effects were reported.

In a second study (Kyriakis et al., 2002), twenty-four crossbred sows (Large White x Landrace) were divided into two equal groups, the experimental group being offered feed incorporating 2% clinoptilolite. The study was maintained for a complete reproductive cycle (day of weaning up to the day of weaning of the next reproductive cycle). Blood samples were collected from each sow on the first day of the study, on days 30 and 90 of pregnancy, at parturition and at weaning. Serum parameters measured included vitamins A and E, inorganic phosphorus, potassium, copper and zinc. The dietary use of clinoptilolite at the inclusion rate of 2% did not provoke any adverse effect on sows/gilts health status and did not provoke any interactive effect on the availability of the dietary mineral elements and vitamins, although the level of vitamin E was found to be slightly lower in the experimental group.

6.4.7 Supplementary animal safety information

The clinoptilolite used in several of the animal safety studies reviewed above is similar in purity and chemical composition to the Zeocem clinoptilolite which is the subject of this notice (Table 6.3). In addition, the Zeocem clinoptilolite contained in ZeoFeed® products is considered to be safe when used as an anticaking agent according to the criteria set forth in the EC/EU Commission Regulations for the addition of clinoptilolite to animal feed (EC, 1999, 2005; EU, 2013, 2018).

Table 6.3	**Comparison of Zeocem clinoptilolite and clinoptilolite used in animal safety studies**		
Reference	Zeocem [a]	Pond and Yen (1983a,b)[b]	Pavelic et al. (2001)
Animals	Ruminants, swine, poultry and pets	Rats	Mice and rats
Parameter	Composition (%)[c]		
Purity	86 - 92	85 - 87 [d]	85[d]
SiO_2	65.0 - 71.3	63.4 - 63.6	50 - 55
Al_2O_3	11.5 - 13.1	12.2 - 12.6	9.3 - 11.4
CaO	2.7 - 5.2	3.0 - 3.4	13.7 - 17.2
Na_2O	0.2 - 1.3	1.4 - 2.5	0.8 - 1.1
Fe_2O_3	0.7 - 2.1	1.3 - 1.9	2.2 - 2.8
K_2O	2.2 - 3.4	1.1 - 2.3	2.9 - 4.3
MgO	0.6 - 1.2	1.5 - 1.7	0.8 - 1.2
TiO_2	0.1 - 0.3	0.1 - 0.3	0.14 - 0.22
Si:Al (ratio)	4.8 - 5.4	4.0 - 4.1	4.8 - 5.4
Particle size	0.01 - 2.5 mm	NG	1.5 - 3.0 µm

[a] Clinoptilolite sourced from Zeocem's mine in Nižný Hrabovec, Slovak Republic (Appendices 2.1 and 2.2).
[b] Pond et al. (1983b) stated that the composition of the clinoptilolite used in the toxicity study was analyzed by Sheppard and Gude III (1982); the same analytical data is reported in Gude III and Sheppard (1988).
[c] Mineral composition of reported oxides.
[d] Estimated amount; calculation based on dated provided in the respective study.
NG - Not given.

6.5 Consumer Safety

Ordinarily, a GRAS notice will address the human dietary consumption of a component of animal feed because of the potential risk to human health due to consumption of animal products and tissues in which the component could be present. However, as the studies reviewed in Section 6.4.1 make evident, there is no need to determine the estimated daily intake for human consumption for clinoptilolite because the substance is excreted by the animal as part of its normal physiological and digestive processes. No data could be found in the scientific literature that would indicate any potential acute or chronic health risks to humans who have consumed animal products from animals that were fed diets containing clinoptilolite. The use of clinoptilolite as an anticaking agent in animal feed in the EU was authorized over 15 years ago. To date, there have been no reports of any adverse effects on consumer safety.

Moreover, because clinoptilolite is a natural crystalline aluminosilicate mineral of sedimentary origin it belongs to the same category of silicates that are classified as GRAS when used as anticaking agents in animal feeds (Sections 6.1.1 - 6.1.3) and is considered safe for use in animal feeds in Europe, (Sections 6.1.5) no safety concerns for consumers need to be addressed.

6.6 Summary and Conclusions

The data and scientific information presented in this document demonstrate that the clinoptilolite which is the subject of this notice has similar physiochemical properties as those silicates (aluminosilicates) listed to be GRAS when used as anticaking agents.

An extensive review of the scientific literature on feeding studies has shown that animal data for poultry and monogastric, ruminant and laboratory animals fed clinoptilolite at levels of 1% or higher of the diet did not indicate any safety issues that could have a negative impact on the health and performance. Some statistically significant changes that have been reported at the use levels of 2% are usually not adverse and therefore do not raise safety concerns. Furthermore, many data have been reported for substances of different origin and composition. Such data needs to be considered, however, as it is not directly applicable to the clinoptilolite described in Part 2 of this notification.

The data reviewed strongly support the premise that the purity and mineral composition of Zeocem clinoptilolite is similar to the purity and composition of other sources of clinoptilolite used in those past animal feeding and safety trials for which no safety concerns were indicated for animal performance or health.

Thus, based on the information provided in this GRAS notice and the scientific procedures discussed herein, the Notifier submits that the Zeocem clinoptilolite of sedimentary origin contained in ZeoFeed® animal products fully meet the criteria of being GRAS when it is used as an anticaking agent at levels not exceeding 1% of the diet, in accordance with good manufacturing practices.

TAB

Part 7

Part 7

DATA AND INFORMATION USED IN THE NOTICE

Table of Contents

7. DATA AND INFORMATION USED IN THE NOTICE

As stated in Part 6, the data and information used in this GRAS Notice are listed separately according to those documents which are generally available and those which are not. Documents that are generally available are listed below in Section 7.1 using standard bibliographic citations.

Those documents that are not generally available are listed in Section 7.2 using the respective names of the Appendices.

7.1 Documentation Generally Available

21 CFR 25.32(f). Foods, food additives, and color additives (see GRAS environmental exclusions).

21 CFR 182.2122. Subpart C, Anticaking agents. Aluminum calcium silicate.

21 CFR 182.2727. Subpart C, Anticaking agents. Sodium aluminosilicate.

21 CFR 182.2729. Subpart C, Anticaking Agents. Sodium calcium aluminosilicate, hydrated.

21 CFR 184.1210. Calcium oxide.

21 CFR 186.1374. Iron oxides.

12 CFR 509.30. Temporary tolerances for polychlorinated biphenyls (PCB's).

21 CFR 570.30. Eligibility for classification as generally recognized as safe (GRAS).

21 CFR 573.940. Silicon dioxide.

21 CFR 582.80. Trace minerals added to animal feeds.

21 CFR 582.1210. Calcium oxide.

21 CFR 582.2122. Subpart C, Anticaking agents. Aluminum calcium silicate.

21 CFR 582.2727. Subpart C, Anticaking agents. Sodium aluminosilicate.

21 CFR 582.2729. Subpart C, Anticaking Agents. Hydrated sodium calcium aluminosilicate.

AAFCO. 2016. 2016 Official Publication. Association of American Feed Control Officials, Inc. Oxford, Indiana. (632 pages).

Abdela, N. 2016. Sub-acute ruminal acidosis (SARA) and its consequence in dairy cattle. A review of past and recent research at global prospective. Achievements in the Life Sciences 10(2):187-196.

Adamis, Z., Tatrai, E., Honma, K., Six, E. and Ungvary, G. 2000. In vitro and in vivo tests for determination of the pathogenicity of quartz, diatomaceous earth, mordenite and clinoptilolite. Annals of Occupational Hygiene 44(1):67-74.

Adhikari, B., Howes, T., Bhandari, B.R. and Truong, V. 2001. Stickiness in foods. A review of mechanisms and test methods. International Journal of Food Properties 4(1):1-33.

Akkoca, D.B., Yılgın, M., Ural, M., Akçin, H. and Mergen, A. 2013. Hydrothermal and thermal treatment of natural clinoptilolite zeolite from Bigadiç, Turkey: An experimental study. Geochemistry International 51(6):495-504.

Alcala-Canto, Y., Gutierrez-Olvera, L., Gutierrez-Olvera, C. and Sumano-Lopez, H. 2011. Effects of clinoptilolite on Eimeria spp. Infection in sheep. Small Ruminant Research 100(2):184-188.

Alexopoulos, C., Papaioannou, D.S., Fortomaris, P., Kyriakis, C.S., Tserveni-Goussi, A., Yannakopoulos, A. and Kyriakis, S.C. 2007. Experimental study on the effect of in-feed administration of a clinoptilolite-rich tuff on certain biochemical and hematological parameters of growing and fattening pigs. Livestock Science 111(3):230-241.

Alver, B.E. 2013. A comparative adsorption study of C_2H_4 and SO_2 on clinoptilolite-rich tuff: Effect of acid treatment. Journal of Hazardous Materials 262:627-633.

Ames, Jr., L.L. 1960. The cation sieve properties of clinoptilolite. American Mineralogist 45:689-700.

Ates, A. and Hardacre, C. 2012. The effect of various treatment conditions on natural zeolites: Ion exchange, acidic, thermal and steam treatments. Journal of Colloid and Interface Science 372(1):130-140.

ATSDR. 1990a. Toxicological profile for radium. Agency for Toxic Substances and Disease Registry. U.S. Public Health Service, Atlanta.

ATSDR. 1990b. Toxicological profile for thorium. Agency for Toxic Substances and Disease Registry. U.S. Public Health Service, Atlanta.

Auerbach, S.M., Carrado, K.A. and Auerbach P.K. (eds). 2003. Handbook of Zeolite Science and Technology. Marcel Dekker Inc., New York, 2003.

Bartko, P., Vrzgula, L., Prosbova, M. and Blazovský, J. 1983a. [The effect of feeding zeolite (clinoptilolite) on the health status of sheep]. Veterinarni Medicina 28(8):481-492. [Abstract].

Bartko, P., Chabada, J., Vrzgula, L., Solar, I. and Blazovsky, J. 1983b. [The fortification of pig feed ration with zeolite in the cage rearing system]. Veterinarni Medicina 28(7):429-435.

Bartko, P., Seidel, H. and Kovac, G. 1995. Use of clinoptilolite-rich tuffs from Slovakia in animal production: A review. pp. 467-475 in: A review: in Natural Zeolites '93: Occurance, Properties, Use. D.W. Ming and F.A. Mumpton (eds). International Committee on Natural Zeolites, Brockport, NY.

Bellia, J.P., Birchall, J.D. and Roberts, N.B. 1996. The role of silicic acid in the renal excretion of aluminium. Annals of Clinical & Laboratory Science 26(3):227-233.

Beltcheva, M., Metcheva, R., Popov, N., Teodorova, S.E., Heredia-Rojas, J.A., Rodríguez-de la Fuente, A.O., Rodríguez-Flores, L.E. and Topashka-Ancheva, M. 2012. Modified natural clinoptilolite detoxifies small mammal's organism loaded with lead I. Lead disposition and kinetic model for lead bioaccumulation. Biological Trace Element Research 147(1-3):180-188.

Bintaş, E., Bozkurt, M., Küçükyılmaz, K., Konak, R., Çınar, M., Akşit, H., Seyrek, K. and Çatlı, A.U. 2014. Efficacy of supplemental natural zeolite in broiler chickens subjected to dietary calcium deficiency. Italian Journal of Animal Science 13(2):275-283

Birchall, J.D., 1992. The interrelationship between silicon and aluminium in the biological effects of aluminium. In Ciba Found Symp (Vol. 169, pp. 50-68).

Birchall, J.D. 1995. The essentiality of silicon in biology. Chemical Society Reviews 24(5):351-357.

Bosi, P., Creston, D. and Casini, L. 2002. Production performance of dairy cows after the dietary addition of clinoptilolite. Italian Journal of Animal Science 1(3):187-195.

Bouda, J. and Jagoš, P. 2014. Biochemical and hematological reference values in calves and their significance for health control. Acta Veterinaria Brno 53(3-4):137-142.

Brun-Hansen, H.C., Kampen, A.H. and Lund, A., 2006. Hematologic values in calves during the first 6 months of life. Veterinary Clinical Pathology 35(2):182-187.

Cerri, G., de' Gennaro, M., Bonferoni, M.C. and Caramella, C. 2004. Zeolites in biomedical application: Zn-exchanged clinoptilolite-rich rock as active carrier for antibiotics in anti-acne topical therapy. Applied Clay Science 27:141-150.

Ceyhan, T., Tatlier, M. and Akçakaya, H. 2007. In vitro evaluation of the use of zeolites as biomaterials: effects on simulated body fluid and two types of cells. Journal of Materials Science: Materials in Medicine 18(8):1557-1562.

CODEX. 2016. General Standard for Food Additives (GSFA) Online. Food Additive Index. FAO/WHO Food Standards. <http://www.fao.org/gsfaonline/additives/index.html>. [Accessed September 2016].

Colella, C. 2011. A critical reconsideration of biomedical and veterinary applications of natural zeolites. Clay Minerals 46(2):295-309.

Coombs, D.S., Alberti, A., Armbruster, T., Artioli, G., Colella, C., Galli, E., Grice, J.D., Liebau, F., Mandarino, J.A., Minato, H., Nickel, E.H., Passaglia, E., Peacor, D.R., Quartieri, S., Rinaldi, R., Ross, M., Sheppard, R.A., Tillmanns, E., and Vezzalini, G. 1997. Recommended nomenclature for zeolite minerals: Report of the Subcommittee on Zeolites of the International Mineralogical Association, Commission on New Minerals and Mineral Names. Canadian Mineralogist 35:1571-1606.

Cotton, A. 2008. Dissolution kinetics of clinoptilolite and heulandite in alkaline conditions. Bioscience Horizons 1(1):38-43.

CPG 7119.14. Guidance Levels for Radionuclides in Domestic and Imported Foods (CPG 7119.14).

CRL. 2010. CRL Evaluation Report on the Analytical Methods submitted in connection with the Application for Authorisation as a Feed Additive according to Regulation (EC) No 1831/2003. Dossier related to: FAD-2009-0017 CRL/09001. Clinoptilolite of sedimentary origin (E 568) European Commission Joint Research Centre, Community Reference Laboratory for Feed Additives. JRC.DG.D.6/CvH/CMP/hn/ARES (2010)-425581. CRL Feed Additives.

Davenward S., Bentham P., Wright J., Crome P., Job D., Polwart A. and Exley C. 2013. Silicon-rich mineral water as a non-invasive test of the 'aluminium hypothesis' in Alzheimer's disease. Journal of Alzheimers Disease 33:423-430.

Deligiannis, K., Lainas, T., Arsenos, G., Papadopoulos, E., Fortomaris, P., Kufidis, D., Stamataris, C. and Zygoyiannis, D. 2005. The effect of feeding clinoptilolite on food intake and performance of growing lambs infected or not with gastrointestinal nematodes. Livestock Production Science 96(2):195-203.

Demirel, R., Yokus, B., Demirel, D., Ketani, A. and Baran, M. 2011. Effects of dietary zeolite on serum contents and feeding performance in rats. International Journal of Agriculture and Biology 13(3):346-350.

Demirel, D.Ş., Demirel, R., Ketani, M.A. and Balcı, K. 2015. Effects on performance, skin and liver histology of different clinoptilolite levels in rat diets. Turkish Journal of Agriculture-Food Science and Technology 3(3):133-141.

Demirel, R., Baran, M.S., DemireL, D.Ş., Ketani, A. and Yokuş, B. 2018. Effects of clinoptilolite on the digestibility of nutrients and relative organ weights in rat diets. Journal of Agricultural Sciences 24(3):297-302.

Domingo, J.L., Gómez, M. and Colomina, M.T. 2011. Oral silicon supplementation: an effective therapy for preventing oral aluminum absorption and retention in mammals. Nutrition Reviews, 69(1):41-51.

Domaracká, L., Štrba, L., Kršák, B. and Muchová, M. 2015. Possiblities of profitable business with nature zeolite – an example from the Nižný Hrabovec deposit, Slovakia. International Journal of Scientific Research and Innovative Technology 2(1):12-19.

Dong, F., Deng, J., Pu, X. and John, H. 2006. [Pulmonary alveolar macrophage cytotoxicity investigation of irregular shape mineral dusts]. Journal of Biomedical Engineering 23(4):848-851.

Dwyer, M.R., Kubena, L.F., Harvey, R.B., Mayura, K., Sarr, A.B., Buckley, S., Bailey, R.H. and Phillips, T.D. 1997. Effects of inorganic adsorbents and cyclopiazonic acid in broiler chickens. Poultry Science 76(8):1141-1149.

EC. 1999. Commission Regulation (EC) No 1245/1999 of 16 June 1999 concerning the authorisation of new additives in feedingstuffs. Official Journal of the European Communites No. L150:15-16, 17.6.199.

EC. 2002. Directive 2002/32/EC of the European Parliament and of the Council of 7 May 2002 on undesirable substances in animal feed. Official Journal of the European Communites No. L140:10-21, 30.5.2002.

EC. 2005. Commission Regulation (EC) No 1810/2005 of 4 November 2005 concerning a new authorisation for 10 years of an additive in feedingstuffs, the permanent authorisation of certain additives in feedingstuffs and the provisional authorisation of new uses of certain additives already authorised in feedingstuffs. Official Journal of the European Communites No. L 291:5-11, 5.11.2005.

EC. 2011. European Commission. Final Report on Feed, EGTOP/1/2011. Directorate-General for Agriculture and Rural Development. Expert Group for Technical Advice on Organic Production (EGTOP).

EC. 2013. Commission Recommendation of 3 December 2013 on the reduction of the presence of dioxins, furans and PCBs in feed and food (2013/711/EU). Official Journal of the European Communites No. L 323.37-39, 4.12.2013.

Edwardson, J.A., Moore, P.B., Ferrier, I.N., Lilley, J.S., Barker, J., Templar, J. and Day, J.P. 1993. Effect of silicon on gastrointestinal absorption of aluminium. The Lancet 342(8865):211-212.

EFSA. 2012a. Scientific Report of EFSA Update of the monitoring of levels of dioxins and PCBs in food and feed. EFSA Journal 10(7):2832 [82 pp.]

EFSA. 2012b. Scientific Opinion on the safety and efficacy of bentonite as a technological feed additive for all species. EFSA Journal 2012;10(7):2787. [20 pp.]

EFSA. 2013. Scientific Opinion on the safety and efficacy of clinoptilolite of sedimentary origin for all animal species. EFSA Journal 11(1):3039-1353.

EFSA. 2016. Scientific opinion on the safety and efficacy of iron oxide black, red and yellow. EFSA Journal 2016 14(6):4482-4495.

Eleroğlu, H., Yalçın, H. and Yıldırım, A. 2011. Dietary effects of Ca-zeolite supplementation on some blood and tibial bone characteristics of broilers. South African Journal of Animal Science 41(4):319-330.

Elliot, M.A. and Edwards JR., H.M. 1991. Comparison of the effects of synthetic and natural zeolite on laying hen and broiler chicken performance. Poultry Science 70(10):2115-2130.

Elmore, A.R. 2003. Final report on the safety assessment of aluminum silicate, calcium silicate, magnesium aluminum silicate, magnesium silicate, magnesium trisilicate, sodium magnesium silicate, zirconium silicate, attapulgite, bentonite, Fuller's earth, hectorite, kaolin, lithium magnesium silicate, lithium magnesium sodium silicate, montmorillonite, pyrophyllite, and zeolite. International Journal of Toxicology 22:37-102.

Enemark, J.M., Frandsen, A.M., Thilsing-Hansen, T. and Jørgensen, R.J., 2003. Aspects of physiological effects of sodium zeolite A supplementation in dry, non-pregnant dairy cows fed grass silage. Acta Veterinaria Scandinavica. Supplementum 97:97-117.

Enemark, Jörg M.D. 2008. The monitoring, prevention and treatment of sub-acute ruminal acidosis (SARA). A review. The Veterinary Journal 176(1):32-43.

EU. 2011. Commission Regulation (EU) No 1129/2011 of 11 November 2011 amending Annex II to Regulation (EC) No 1333/2008 of the European Parliament and of the Council by establishing a Union list of food additives. Official Journal of the European Communities No. L 295:1-177, 12.11.2011.

EU. 2013. Commission Implementing Regulation (EU) No 651/2013 of 9 July 2013 concerning the authorisation of clinoptilolite of sedimentary origin as a feed additive for all animal species and amending Regulation (EC) No 1810/2005. Official Journal of the European Communities L 189:1-3, 10.7.2013.

EU. 2018. European Union Register of Feed Additives pursuant to Regulation (EC) No 1831/2003. Annex I: List of additives. Edition 4/2018 (264) (Released 03.08.2018). Directorate - General for Health and Food Safety, Directorate E - Veterinary and International Affairs, Unit E5 Animal Nutrition. <https://ec.europa.eu/food/sites/food/files/safety/docs/animal-feed-eu-reg-comm_register_feed_additives_1831-03.pdf>. [Accessed October 2018].

Farias, T., Ruiz-Salvador, A.R. and Rivera, A., 2003. Interaction studies between drugs and a purified natural clinoptilolite. Microporous and Mesoporous Materials 61(1-3):117-125.

FAO. 2005a. Calcium aluminium silicate. Combined Compendium of Food Additive Specifications: Volume 1 - Food Additives A-D. FAO JECFA Monographs 1. pp. 205-207. Food and Agriculture Organization, Rome.

FAO. 2005b. Sodium aluminosilicate. Combined Compendium of Food Additive Specifications: Volume 1 - Food Additives P-Z. FAO JECFA Monographs 1. pp. 305-208. Food and Agriculture Organization, Rome.

FCC. 2010. Food Chemicals Codex 2010-2011. Seventh Edition. U.S. Pharmacopeial Convention, Rockville, MD. pp. 921-923.

FDA. 1995. CVM's Compliance Policy Guide CPG 665.100 (Common or Usual Names for Animal Feed Ingredients). <http://www.fda.gov/ICECI/ComplianceManuals/CompliancePolicyGuidanceManual /ucm074687.htm>. [Accessed January 2016].

FDA. 2005. Guidance Levels for Radionuclides in Domestic and Imported Foods (CPG 7119.14).

FDA. 2015. Select Committee on GRAS Substances (SCOGS) Opinion: Silicates. U.S. Food and Drug Administration, Silver Spring, MD. <http://www.fda.gov/Food/ IngredientsPackagingLabeling /GRAS/SCOGS/ucm260849.htm>. [Accessed January 2016].

FDA. 2016a. Title 21--Food and Drugs, Chapter I--Food and Drug Administration, Department of Health and Human Services, Subchapter B--Food for Human Consumption, Part 182 Substances Generally Recognized As Safe. Subpart C, Anticaking Agents. U.S. Food and Drug Administration, Silver Spring, MD.

FDA. 2016b. Title 21--Food and Drugs, Chapter I--Food and Drug Administration, Department of Health and Human Services, Subchapter E--Animal Drugs, Feeds, and Related Products, Part 582 Substances Generally Recognized As Safe. Subpart C, Anticaking Agents. U.S. Food and Drug Administration, Silver Spring, MD.

FDA. 2016c. Animal and Veterinary, FDA's Regulation of Pet Food, Resources for You, Generally Recognized as Safe. U.S. Food and Drug Administration, Silver Spring, MD. <http:// www.fda.gov/animalveterinary/resourcesforyou/ucm047111 .htm>. [Accessed January 2016].

FEDIAF. 2013. Fédération européenne de lindustrie des aliments pour animaux familiers (European Pet Food Industry Federation). Nutritional Guidelines: For Complete and Complementary Pet Food for Cats and Dogs. European Pet Food Industry, Brussels. 85 pp.

Fokas, P., Zervas, G., Fegeros, K. and Zoiopoulos, P. 2004. Assessment of Pb retention coefficient and nutrient utilization in growing pigs fed diets with added clinoptilolite. Animal Feed Science and Technology 117(1):121-129.

Gerasev, A.D., Lukanina, S.N., Svyatash, G.A. and Aizman, R.I. 2003a. K^+ Transport in the rat gut following natural zeolites intake as a food supplement. Russian Physiological Journal 89(8):972-981.

Gerasev, A.D., Svyatash, G.A., Lukanina, S.N., Taranov, A.G. and Aizman, R.I. 2003b. Effects of natural Zeolites on renal function and water-salt metabolism in rats. Russian Journal of Physiology 89 (7):879-887.

Gezen, S.S., Eren, M., Balcı, F., Deniz, G., Biricik, H. and Bozan, B. 2009. The effect of clinoptilolite in low calcium diets on performance and eggshell quality parameters of aged hens. Asian-Australasian Journal of Animal Sciences 22(9):1296-1302.

GPO. 1958. Public Law 85-929 [72 Stat.] An Act. September 6, 1958. [H.R. 13254]. Food Additives Amendment of 1958. 52 Stat. 1041. 21 USC 321. Definitions.

GPO. 2016a. Federal Register, Vol. 81, No. 159, Wednesday, August 17, 2016, Part III, Department of Health and Human Services, Food and Drug Administration, 21 CFR Parts 20, 25, 170, et al., Substances Generally Recognized as Safe: Final Rule. U.S. Government Publishing Office, Washington, DC.

GPO. 2016b. Title 21: Food and Drugs, Part 582 - Substances generally recognized as safe. Subpart C - Anticaking Agents. U.S. Government Publishing Office, Washington, DC.

Gude III, A.J. and Sheppard, R.A. 1988. A zeolitic tuff in a lacustrine facies of the Gila Conglomerate near Buckhorn, Grant County, New Mexico. US Geological Survey Bulletin 1763.

Herzig, I., Strakova, E. and Suchy, P. 2008. Long-term application of clinoptilolite via the feed of layers and its impact on the chemical composition of long bones of pelvic limb (femur and tibiotarsus) and eggshell. Veterinarni Medicina 53(10): 550-554.

Hollenbach, A.M., Peleg, M. and Rufner, R. 1982. Effect of four anticaking agents on the bulk characteristics of ground sugar. Journal of Food Science 47(2):538-544.

Hutcheson, DE. 1984. Addition of clinoptilolite ores to the diets of feeder cattle, pp. 195-199. In: W.G. Pond and F.A. Mumpton (eds). Zeo-agriculture: Use of Natural Zeolites in Agriculture and Aquaculture. Westview Press, Inc. Boulder, CO. [cited in Katsoulos, P.D et al (2005a)].

IDALS. 1999. Mycotoxin binding claims on anticaking agents. Commercial Feed and Fertilizer Bureau, Iowa Department of Agriculture and Land Stewardship. Des Moines, IA.

Inglezakis, V.J. 2005. The concept of "capacity" in zeolite ion-exchange systems. Journal of Colloid and Interface Science 281(1):68-79.

Institute of Medicine. 2003. Dioxins and Dioxin-like Compounds in the Food Supply: Strategies to Decrease Exposure. Washington, DC: The National Academies Press. <https://www.ncbi.nlm.nih.gov/books/NBK221718/>. Accessed August 2018.

Irani, R.R., Callis, C.F. and Liu, T. 1959. Flow conditioning anticaking agents. Industrial & Engineering Chemistry Research 51(10):1285-1288.

Irani, R.R., Vandersall, H.L. and Morgenthaler, W.W. 1961. Water vapor sorption in flow conditioning and cake inhibition. Industrial & Engineering Chemistry Research 53(2):141-142.

Jenike, A.W. 1964. Storage and flow of solids. Bulletin of the University of Utah, Vol. 53, No. 26. Bulletin No. 123 of the Utah Engineering Experimental Station. Sixth Printing (revised), March 1970. University of Utah, Salt Lake, Utah.

Jha, V.K. and Hayashi, S. 2009. Modification on natural clinoptilolite zeolite for its NH 4+ retention capacity. Journal of Hazardous Materials, 169(1):29-35.

Jones, K., Linhart, C., Hawkins, C. and Exley, C. 2017. Urinary excretion of aluminium and silicon in secondary progressive multiple sclerosis. EBioMedicine 26:60-67.

Jorgensen, T.C. and Weatherley, L.R. 2003. Ammonia removal from wastewater by ion exchange in the presence of organic contaminants. Water Research 37(8):1723-1728.

Kadirbekov, K., Zhambakin, D., Kadirbekov, A. and Imanbekov, K. 2017. Acid activation of natural zeolite with high content of iron oxides in creation of selective sorbents and catalysts. In MATEC Web of Conferences 96:00002. EDP Sciences.

Karamanlis, X., Fortomaris, P., Arsenos, G., Dosis, I., Papaioannou, D., Batzios, C., and Kamarianos, A. 2008. The effect of a natural zeolite (clinoptilolite) on the performance of broiler chickens and the quality of their litter. Asian-Australian Journal of Animal Science 21: 1642-1650.

Karatzia, M.A., Pourliotis, K., Katsoulos, P.D. and Karatzias, H. 2011. Effects of in-feed inclusion of clinoptilolite on blood serum concentrations of aluminium and inorganic phosphorus and on ruminal pH and volatile fatty acid concentrations in dairy cows. Biological Trace Element Research 142(2):159-166.

Karatzia, M.A., Katsoulos, P.D. and Karatzias, H. 2013. Diet supplementation with clinoptilolite improves energy status, reproductive efficiency and increases milk yield in dairy heifers. Animal Production Science 53(3):234-239.

Katsoulos, P.D., Roubies, N., Panousis, N., Christaki, E., Karatzanos, P. and Karatzias, H. 2005a. Effects of long term feeding dairy cows on a diet supplemented with clinoptilolite on certain haematological parameters. Veterinarni Medicina 50(10):427-431.

Katsoulos, P.D., Panousis, N., Roubies, N., Christaki, E. and Karatzias, H. 2005b. Effects on blood concentrations of certain serum fat-soluble vitamins of long-term feeding of dairy cows on a diet supplemented with clinoptilolite. Journal of Veterinary Medicine Series A, 52(4):157-161.

Katsoulos, P.D., Roubies, N., Panousis, N., Arsenos, G., Christaki, E. and Karatzias, H. 2005c. Effects of long-term dietary supplementation with clinoptilolite on incidence of parturient paresis and serum concentrations of total calcium, phosphate, magnesium, potassium, and sodium in dairy cows. American Journal of Veterinary Research 66(12):2081-2085.

Katsoulos, P.D., Roubies, N., Panousis, N. and Karatzias, H. 2005d. Effects of long-term feeding dairy cows on a diet supplemented with clinoptilolite on certain serum trace elements. Biological Trace Element Research 108(1-3):137-145.

Katsoulos, P.D., Panousis, N., Roubies, N., Christaki, E., Arsenos, G. and Karatzias, H. 2006. Effects of long-term feeding of a diet supplemented with clinoptilolite to dairy cows on the incidence of ketosis, milk yield and liver function. Veterinary Record: Journal of the British Veterinary Association 159(13):415-418.

Katsoulos, P.D., Zarogiannis, S., Roubies, N. and Christodoulopoulos, G. 2009. Effect of long-term dietary supplementation with clinoptilolite on performance and selected serum biochemical values in dairy goats. American Journal of Veterinary Research 70(3):346-352.

Katsoulos, P.D., Karatzia, M.A., Polizopoulou, Z., Florou-Paneri, P. and Karatzias, H. 2017. Effects of prolonged consumption of water with elevated nitrate levels on certain metabolic parameters of dairy cattle and use of clinoptilolite for their amelioration. Journal of Dairy and Veterinary Sciences, 1(2):1-5.

Kavak, D.D. and Ülkü, S. 2012. Investigation of structural properties of clinoptilolite rich zeolites in simulated digestion conditions and their cytotoxicity against Caco-2 cells in vitro. Journal of Porous Materials 20(2):331-338.

Kavan, B.P., Shargh, M.S., Hassani, S. and Mostafalo, Y. 2013. Comparison of the effects of clinoptilolite and sodium zeolite A on tibia bone mineralisation and calcium and phosphorus utilisation in broiler chicks. International Research Journal of Applied and Basic Sciences 4(11): 3389-3395.

Kaya, H., Kaya, A., Gul, M. and Celebi, S. 2013. Effect of zeolite and organic acid mixture supplementation in the layer diet on performance, egg quality traits and some blood parameters. Journal of Animal and Veterinary Advances 12(6):782-787.

Kithome, M., Paul, J.W., Lavkulich, L.M., and Bomke, A.A. 1999 Effect of pH on ammonium adsorption by natural zeolite clinoptilolite. Commun. Soil. Sci. Plan, 30 (9–10): 1417–1430.

Klinkon, M. and Ježek, J. 2012. Values of blood variables in calves. In A Bird's-Eye View of Veterinary Medicine. <www.intechopen.com>.

Kotova, D.L., Artamonova, M.N., Krysanova, T.A., Novikova, L.A. and Belchinskaya, L.I., 2016. Adsorption of water by clinoptilolite and glauconite. Sorption and chromatography processes. 16(3):390-395.

Kovac, G., Bartko, P., Vrzgula, L., Reichel, P., Nistiari, F., Mojzis, J. and Seidel, H. 1995. Application of clinoptiloite during organophosphate intoxication in sheep, pp. 459-466. In: Natural Zeolites '93: Occurrence, Properties, Use. D.W. Ming and F.A. Mumpton (eds). International Committee on Natural Zeolites, Brockport, NY.

Kralik, G., Šperanda, M. and Gajčević, Z. 2006, January. Effects of Nanofeed® preparation on egg quality and biochemical indicators in laying hens' serum. In Proceedings of the XII European Poulty Conference, Verona, Italy, Vol. 196:1-6.

Kraljević Pavelić, S. Micek, V., Filošević, A., Gumbarević, D., Žurga, P., Bulog, A., Orct, T., Yamamoto, Y., Preočanin, T., Plavec, J. and Peter, R., 2017. Novel, oxygenated clinoptilolite material efficiently removes aluminium from aluminium chloride-intoxicated rats in vivo. Microporous and Mesoporous Materials 249:146-156.

Krause, K.M. and Oetzel, G.R. 2006. Understanding and preventing subacute ruminal acidosis in dairy herds. A review. Animal Feed Science and Technology 126(3-4):215-236.

Krewski, D., Yokel, R.A., Nieboer, E., Borchelt, D., Cohen, J., Harry, J., Kacew, S., Lindsay, J., Mahfouz, A.M. and Rondeau, V., 2007. Human health risk assessment for aluminium, aluminium oxide, and aluminium hydroxide. Journal of Toxicology and Environmental Health, Part B, 10(S1):1-269.

Kyriakis, S.C., Papaioannou, D.S., Alexopoulos, C., Polizopoulou, Z., Tzika, E.D. and Kyriakis, C.S. 2002. Experimental studies on safety and efficacy of the dietary use of a clinoptilolite-rich tuff in sows: a review of recent research in Greece. Microporous and Mesoporous Materials 51(1):65-74.

Lam, A., Rivera, A. and Rodrídguez-Fuentes, G. 2001. Theoretical study of metronidazole adsorption on clinoptilolite. Microporous and Mesoporous Materials 49(1-3):157-162.

Lam, A., Sierra, L.R., Rojas, G., Rivera, A., Rodrıguez-Fuentes, G. and Montero, L.A. 1998. Theoeretical study of the physical adsorption of aspirin on natural clinoptilolite. Microporous and Mesoporous Materials 23(5-6):247-252.

Leung, S., Barrington, S., Wan, Y., Zhao, X. and El-Husseini, B. 2007. Zeolite (clinoptilolite) as feed additive to reduce manure mineral content. Bioresource Technology 98(17):3309-3316.

Li, L.Y., Tazaki, K., Lai, R., Shiraki, K., Asada, R., Watanabe, H. and Chen, M. 2008. Treatment of acid rock drainage by clinoptilolite—adsorptivity and structural stability for different pH environments. Applied Clay Science 39(1):1-9.

LSRO. 1979. Evaluation of the Health Aspects of Certain Silicates as Food Ingredients. SCOGS Report Number: 61, Contract No. FDA 223-75-2004. Federation of American Societies for Experimental Biology, Bethesda, MD. Life Sciences Research Office; Food and Drug Administration, Washington, DC. Bureau of Foods.

Macháček, M., Večerek, V., Mas, N., Suchý, P., Straková, E., Šerman, V. and Herzig, I. 2010. Effect of the feed additive clinoptilolite (ZeoFeed) on nutrient metabolism and production performance of laying hens. Acta Veterinaria Brno 79(9):29-34.

Maia, G.V.C.; Saad, F.M.O.B.; Roque, N.C. *et al.* 2010. Zeólitas e Yucca schidigera em rações para cães: palatabilidade, digestibilidade e redução de odores fecais. Revista Brasileira de Zootecnia 19:2242-2246. [Portuguese, English Abstract].

Margeta, K., Logar, N.Z., Šiljeg, M. and Farkas, A. 2013. Natural zeolites in water treatment–how effective is their use. In Water treatment. InTech. >http://dx.doi.org/10.5772/50738>

Martin-Kleiner, I., Flegar-Meštrić, Z., Zadro, R., Breljak, D., Janda, S. S., Stojković, R., Marušić, M., Radačić, M. and Boranić, M. 2001. The effect of the zeolite clinoptilolite on serum chemistry and hematopoiesis in mice. Food and Chemical Toxicology 39(7):717-727.

McCollum, F.T. and Galyean, M.L. 1983. Effects of clinoptilolite on rumen fermentation, digestion and feedlot performance in beef steers fed high concentrate diets. Journal of Animal Science 56(3):517-524.

Miles, R.D. and Henry, P.R. 2007. Safety of improved Milbond-TX® when fed in broiler diets at greater than recommended levels. Animal Feed Science and Technology 138(3):309-317.

Minceva, M., Taparcevska, J., Markovska, L., Koumanova, B. and Meshko, V. 2008. Adsorption kinetics of Pb2+ onto natural zeolite. Journal of the University of Chemical Technology and Metallurgy 43(1):93-100.

Moghaddam, H.N., Jahanian, R., Najafabadi, H.J. and Madaeni, M.M. 2008. Influence of dietary zeolite supplementation on the performance and egg quality of laying hens fed varying levels of calcium and nonphytate phosphorus. Journal of Biological Sciences 8(2):328-334.

Mohri, M., Seifi, H.A. and Maleki, M. 2008a. Effects of short-term supplementation of clinoptilolite in colostrum and milk on the concentration of some serum minerals in neonatal dairy calves. Biological Trace Element Research 123(1-3):116-123.

Mohri, M., Seifi, H.A. and Daraei, F. 2008b. Effects of short-term supplementation of clinoptilolite in colostrum and milk on hematology, serum proteins, performance, and health in neonatal dairy calves. Food and Chemical Toxicology 46(6):2112-2117.

Mojzis J, Nistiar F, Kovác G and Mojzisová G. 2008. Effects of short-term supplementation of clinoptilolite in colostrum and milk on the concentration of some serum minerals in neonatal dairy calves. Biological Trace Element Research 123(1-3):116-123.

Mumpton, F.A. 1960. Clinoptilolite redefined. American Mineralogist 45:351-369.

Mumpton, F.A. 1985. Using zeolites in agriculture, pp. 127-158. In: Innovative Biological Technologies for Lesser Developed Countries, Washington, DC: US Congress, Office of Technology Assessment, OTA-13P-F-29. 1985.

Mumpton, F.A. 1999. La roca magica: uses of natural zeolites in agriculture and industry. Proceedings of the National Academy of Sciences 96(7):3463-3470.

Mumpton, F.A. and Fishman, P.H. 1977. The application of natural zeolites in animal science and aquaculture. Journal of Animal Science 45(5):1188-1203.

Nakaue, H.S. and Koelliker, J.K. 1981. Studies with clinoptilolite in poultry I. Effect of feeding varying levels of clinoptilolite (zeolite) to dwarf single comb white leghorn pullets and ammonia production. Poultry Science 60(5):944-949.

NRC. 1980. Mineral tolerance of domestic animals. National Research Council. U.S. Subcommittee on Mineral Toxicity in Animals. National Academies Press, Washington, D.C.

NRC. 1994. Nutrient Requirements of Poultry, Ninth Revised Edition. National Research Council, National Academy Press, Washington, D.C.

NRC. 1998. Nutrient Requirements of Swine, Tenth Revised Edition. National Research Council, National Academy Press, Washington, D.C.

NRC. 2000. Nutrient Requirements of Beef Cattle, Seventh Revised Edition. National Research Council, National Academy of Sciences, Washington, D.C.

NRC. 2001. Nutrient Requirements of Dairy Cattle, Seventh Revised Edition. National Research Council, National Academy of Sciences, Washington, D.C.

NRC. 2005. Mineral Tolerance of Animals, Second Revised Edition. National Research Council. The National Academies Press, Washington, DC.

NRC. 2006. Nutrient Requirements of Dogs and Cats, National Research Council, National Academy of Sciences, Washington, D.C.

NRC. 2007. Nutrient Requirements of Small Ruminants. National Research Council. The National Academies Press, Washington, D.C.

Nestorov, N. 1984. Possible applications of natural zeolites in animal husbandry, pp. 163-169 In: Natural Zeolites '93: Occurrence, Properties, Use. D.W. Ming and F.A. Mumpton (eds). International Committee on Natural Zeolites, Brockport, NY.

Nezamzadeh-Ejhieh, A. and Tavakoli-Ghinani, S. 2014. Effect of a nano-sized natural clinoptilolite modified by the hexadecyltrimethyl ammonium surfactant on cephalexin drug delivery. Comptes Rendus Chimie 17(1):49-61.

Norouzian, M.A., Valizadeh, R.E.Z.A., Khadem, A.A., Afzalzadeh, A. and Nabipour, A. 2010. The effects of feeding clinoptilolite on hematology, performance, and health of newborn lambs. Biological Trace Element Research 137(2):168-176.

Oğuz, H. and Kurtoğlu, V. 2000. Effect of clinoptilolite on performance of broiler chickens during experimental aflatoxicosis. British Poultry Science 41(4):512-517.

Oğuz, H., Kurtoğlu, F., Kurtoğlu, V. and Bırdane, Y.O. 2002. Evaluation of biochemical characters of broiler chickens during dietary aflatoxin (50 and 100 ppb) and clinoptilolite exposure. Research in Veterinary Science 73(1):101-103.

OLRC. 2016. Federal Food and Drug Cosmetic Act, Chapter III, Sec. 301 [21 U.S.C. 331] (II)(3)(C) [As Amended Through P.L. 114-146, Enacted April 19, 2016] (OLRC, 2016).

Olver, M.D. 1989. Effect of feeding clinoptilolite (zeolite) to three strains of laying hens. British Poultry Science, 30(1), 115-121.

Ortatatli, M., Oğuz, H., Hatipoğlu, F. and Karaman, M. 2005. Evaluation of pathological changes in broilers during chronic aflatoxin (50 and 100 ppb) and clinoptilolite exposure. Research in Veterinary Science 78(1):61-68.

Öztürk, E., Erener, G. and Sarica, M. 1998. Influence of natural zeolite on performance of laying hens and egg quality. Turkish Journal of Agriculture and Forestry 22(6):623-628.

Papaioannou, D.S., Kyriakis, S.C., Papasteriadis, A., Roumbies, N., Yannakopoulos, A. and Alexopoulos, C. 2002. Effect of in-feed inclusion of a natural zeolite (clinoptilolite) on certain vitamin, macro and trace element concentrations in the blood, liver and kidney tissues of sows. Research in Veterinary Science 72(1):61-68.

Papaioannou, D., Katsoulos, P.D., Panousis, N. and Karatzias, H. 2005. The role of natural and synthetic zeolites as feed additives on the prevention and/or the treatment of certain farm animal diseases: a review. Microporous and Mesoporous Materials 84(1):161-170.

Parizadian, K. 2013. Effects of physical size of clinoptilolite on growth performance, serum biochemical parameters and litter quality of broiler chickens in the growing phase. Poultry Science Journal 1(2):93-104.

Parlat, S.S., Yildiz, A.O. and Oguz, H. 1999. Effect of clinoptilolite on performance of Japanese quail (Coturnix coturnix japonica) during experimental aflatoxicosis. British Poultry Science 40(4):495-500.

Pavelić, K., Hadzija, M., Bedrica, L., Pavelić, J., Dikić, I., Katić, M. et al. 2001. Natural zeolite clinoptilolite: new adjuvant in anticancer therapy. Journal of Molecular Medicine 78(12):708-720.

Peleg, M. and Hollenbach, A.M. 1984. Flow conditioners and anticaking agents. Recent Advances in Engineering Properties of Foods 3:93-102.

Polatoğlu, İ. 2005. Chemical behaviour of clinoptilolite rich natural zeolite in aqueous medium. Master Thesis, İzmir Institute of Technology. Izmir Turkey. 88 pp.

Pond, W.G. and Yen, J.T. 1983a. Protection by clinoptilolite or zeolite NaA against cadmium-induced anemia in growing swine. Experimental Biology and Medicine 173(3):332-337.

Pond W.G and Yen J.T 1983b Reproduction and progeny growth in rats fed clioptilolite in the presence or absence of dietary cadmium. Bulletin of Environmental Contamination and Toxicology 31:666-672.

Pond, W.G. and Yen, J.T., 1984. Physiological effects of clinoptilolite and synthetic zeolite A, pp. 127-142. In: W.G. Pond and F.A. Mumpton (eds). Zeo-agriculture: Use of Natural Zeolites in Agriculture and Aquaculture. Westview Press, Inc. Boulder, CO.

Pond, W.G., Laurent, S.M. and Orloff, H.D. 1984. Effect of dietary clinoptilolite or zeolite Na-A on body weight gain and feed utilization of growing lambs fed urea or intact protein as a nitrogen supplement. Zeolites 4(2):127-132.

Pond, W.G., Yen, J.T., and Varel, V.H. 1988. Response of growing swine to dietary copper and clinoptilolite supplementation. Nutrition Reports International 4:795-803.

Pond, W.G., Yen, J.T. and Crouse, J.D. 1989. Tissue mineral element content in swine fed clinoptilolite. Bulletin of Environmental Contamination and Toxicology 42(5):735-742.

Poulsen, H. and Oksbjerg, N. 1995. Effects of dietary inclusion of a zeolite (clinoptilolite) on performance and protein metabolism of young growing pigs. Animal Feed Science and Technology 53(3-4): 297-303.

Pourliotis, K., Karatzia, M.A., Florou-Paneri, P., Katsoulos, P.D. and Karatzias, H. 2012. Effects of dietary inclusion of clinoptilolite in colostrum and milk of dairy calves on absorption of antibodies against Escherichia coli and the incidence of diarrhea. Animal Feed Science and Technology 172(3):136-140.

Prvulović, D., Jovanović-Galović, A., Stanić, B., Popović, M. and Grubor-Lajšić, G. 2007. Effects of a clinoptilolite supplement in pig diets on performance and serum parameters. Czech Journal of Animal Science 52(6):159-164.

PubChem. 2016. Clinoptilolite. Open Chemistry Database. National Institute of Health, MD. <https://pubchem.ncbi.nlm.nih.gov/compound/6336554#section=Top>. [Accessed January 2016].

Ramos, A.J. and Hernandez, E. 1997. Prevention of aflatoxicosis in farm animals by means of hydrated sodium calcium aluminosilicate addition to feedstuffs: a review. Animal Feed Science and Technology 65(1):197-206.

Reháková, M., Čuvanová, S., Dzivak, M., Rimár, J. and Gaval'ová, Z. 2004. Agricultural and agrochemical uses of natural zeolite of the clinoptilolite type. Current Opinion in Solid State and Materials Science 8(6):397-404.

Rivera, A., Rodrıguez-Fuentes, G. and Altshuler, E. 1998. Characterization and neutralizing properties of a natural zeolite/Na_2CO_3 composite material. Microporous and Mesoporous Materials 24(1):51-58.

Rivera, A., Farías, T., de Ménorval, L.C., Autié-Pérez, M. and Lam, A. 2013. Natural and sodium clinoptilolites submitted to acid treatments: Experimental and theoretical studies. The Journal of Physical Chemistry C 117(8):4079-4088.

Roque, N.C., Saad, F.M.D.O.B., Santos, J.P.F.D., Ebina, F.S., Chizzotti, A.F., Silva, R.C., Aquino, A.A. and Maia, G.V.C. 2011. Increasing levels of zeolite and Yucca schidigera in diets for adult cats. Revista Brasileira de Zootecnia 40(11):2471-2475.

Ruíz-Barrera, O., Castillo-Castillo, Y., Arzola-Álvarez, C. and Rodriguez-Muela, C. 2006. Effects of graded levels of Zeolite on digestibility and nutrient intake of sheep. In Proceedings-American Society of Animal Science Western Section 57:322-324.

Sacakli, P., Calik, A., Bayraktaroglu, A.G., Ergun, A., Sahan. O. and Ozaydin, S. 2015. AG, B., 2015. Effect of clinoptilolite and/or phytase on broiler growth performance, carcass characteristics, intestinal histomorphology and tibia calcium and phosphorus levels. Kafkas Üniversitesi Veteriner Fakültesi Dergisi 21(5):729-737).

Safameher, A. 2008. Effects of clinoptilolite on performance, biochemical parameters and hepatic lesions in broiler chickens during aflatoxicosis. Journal of Animal and Veterinary Advances 7(4):381-388.

Šamanc, H., Kirovski, D., Adamoviš, M., Vujanaš, I., Fratriš, N. and Prodanoviš, R. 2008. Effects of natural zeolite on body weight, weight gain, hematology and biochemical blood parameters in calves. Veterinarski Glasnik 62:153-166.

Santos, J.P.F., Saad, F.M.O.B., Roque, N.C., Aquino, A.A., Pires, C.P. and Geraldi, L.F. 2011. Yucca schidigera e zeólita em alimento para gatos adultos e seus efeitos na excreção de minerais. Arquivo Brasileiro de Medicina Veterinária e Zootecnia 63(3):687-693. [Portuguese, English Abstract].

Santos, J.P.F., Saad, F.M.O.B., Maia, G.V.C., Pires, C.P., Reis, J.S., Roque, N.C., Aquino, A.A. and Brunetto, M. A. 2013. Inclusion of Yucca schidigera and zeolite in dog feed: influence on mineral excretion. Arquivo Brasileiro de Medicina Veterinária e Zootecnia 65(3):918-926.

Santos, J.P.F., Saad, F.M.O.B., Ogoshi, R. C. S., Reis, J. S. D., Ferreira, L. G., Pires, C.P. Franca, J., Ebina, F.S., Aquino, A.A. and Brunetto, M.A. 2016. Inclusion of Yucca schidigera extract and zeolite in the diet and its relationship to the apparent digestibility of nutrients and urinary pH in adult dogs. Ciência Rural 46(8):1456-1459.

Sardi, L., Martelli, G., Parisini, P., Cessi, E. and Mordenti, A. 2002. The effects of clinoptilolite on piglet and heavy pig production. Italian Journal of Animal Science 1(2):103-112.

Scheideler, S.E. 1993. Effects of various types of aluminosilicates and aflatoxin B_1 on aflatoxin toxicity, chick performance, and mineral status. In Poultry Science 72 (2):282-288.

Sheppard, R.A. 1971. Clinoptilolite of possible economic value in sedimentary deposits of the conterminous United States. Geological Survey Bulletin 1332-B. U.S. Government Printing Office.

Sheppard, R.A. and Gude III, A.J. 1982. Mineralogy, chemistry, gas adsorption, and NH_4 exchange capacity for selected zeolitic tuffs from the Western United States. Open-File Report 82-969. US Geological Survey.

Shurson, G.C., Ku, P.K., Miller, E R. and Yokoyama, M.T. 1984. Effects of zeolite A or clinoptilolite in diets of growing swine. Journal of Animal Science 59(6):1536-1545.

Step, D., Litherland, N., Burciaga-Robles, L., Breshears, M., Krehbiel, C., Confer, A., Fulton, R., Morgan, G., Thornsberry, R., Fassing, S. 2008. Clinical observations, biochemical data, and postmortem and histopathologic findings in young dairy calves fed zeolite clinoptilolite binder combined with milk replacer. American Journal of Veterinary Research 69(12):1587-1594.

Straková, E., Suchy, P., Herzig, I., Serman, V. and Mas, N. 2008. The long-term administration of a clinoptilolite-supplemented feed to layers and its effect on performance, haematological parameters and metabolic profile. Czech Journal of Animal Science 53(5):212.

Subramaniam, M.D. and Kim, I.H. 2015. Clays as dietary supplements for swine: A review. Journal of Animal Science and Biotechnology 6(1):38-46.

Suchý, P., Strakova, E., Večerek, V., Klouda, Z. and Kráčmarová, E. 2006. The effect of a clinoptilolite-based feed supplement on the performance of broiler chickens. Czech Journal of Animal Science 51(4):168-173.

Sweeney, T.F., Cervantes, A., Bull, L.S. and Hemken, R.W. 1983. Effect of dietary clinoptilolite on digestion and rumen fermentation in steers, pp. 183-193. In: Zeo-agriculture: use of natural zeolites in agriculture and aquaculture. Boulder, Colo: Westview Press Inc.

Taparcevska, J., Markovska, L., Koumanova, B. and Meshko, V. 2010. Diffusion models for adsorption kinetics of Zn^{2+}, Cd^{2+} and Pb^{2+} onto natural zeolite. Water Science and Technology 62(5):1136-1142.

Toprak, N.N., Yılmaz, A., Öztürk, E., Yigit, O. and Cedden, F. 2016. Effect of micronized zeolite addition to lamb concentrate feeds on growth performance and some blood chemistry and metabolites. South African Journal of Animal Science 46(3):313-320.

Tátrai, E. and Ungváry, G. 1993. Study on carcinogenicity of clinoptilolite type zeolite in Wistar rats. Polish Journal of Occupational Medicine and Environmental Health 6(1):27-34.

Thomas, J.A. and Ballantyne, B. 1992. Toxicological assessment of zeolites. Journal of the American College of Toxicology 11(3):259-273.

Troszkiy, V., Hnativ, Z., Duleba, V. and Kriuchenko, T. 2017. Investigation of the processes of chemical (ACID) labilizing of clinoptilolite. Environmental Problems 2(2):71-76.

USGS. 2015, Mineral commodity summaries 2015. U.S. Geological Survey, 196p. <http://dx.doi.org/10.3133/70140094>. [Accessed April 2016].

Utlu, N., Celebi, S., Yücel, O. 2007. The Effects of natural zeolite supplementation to diet on serum element concentrations in laying hens. Revue Méd. Vét., 158, 12, 598-602

Vatalová, M., Vida, M., Šamajová, E. and Janotka, I. 2016. Sedimentary Zeolite Deposits in Slovakia. <http://www.iza-online.org/natural/Catalog/Slovakia/SlovakiaFrame.htm>. [Accessed April 2016].

Vrzgula, L. 1985. Natural zeolite/clinoptilolite in the prevention and therapy of calf diarrhoea of alimentary etiology, 1A-8, pp. 365-366. In: Reprints of Posters. 7th International Zeolite Conference, 17-22 August, Tokyo. Japan Association of Zeolites.

Vrzgula, L., Bartko, P., Blazovsky, J. Kozac, J. 1982. [Effect of feeding clinoptilolite on health status, blood count, and weight gains of pigs]. Veterinarni Medicina 27(5):267-274.

Vrzgula, L., Bartko, P., Solar, I and Reichel, P. 1984. [Fortification of the feeding ration by natural zeolite in fattening pigs]. Veterinarni Medicina 28(11):679-686.

Vrzgula, L., Bartko, P., Kovac, G., Prosbova, A., Reichel, P., Paulikoya, I., Bires, J. and Seidel, H. 1985. Clinical and laboratory studies of natural zeolite in cattle and sheep. In: Research Report VSV Kosice, November, 253 pp. (reviewed by Bartko et al., 1995)

Vrzgula, L., Prosbova, M., Blazovsky, J., Jacobi, U., Schubert, T. and Kovac, G. 1988. The effect of feeding natural zeolite on indices of the internal environment of calves in the postnatal period. pp. 747-752 in: Occurrence, Properties and Utilization of Natural Zeolites. D. Kallo and H.S. Sherry (eds). Akademiai Kiado, Budapest.

Waluyo, J., Makertihartha, I.G.B.N. and Susanto, H. 2018. The effect of acid leaching time in modifying natural zeolite as catalyst for toluene steam reforming. In MATEC Web of Conferences 159:02060.

WHO. 1984. Evaluation of certain food additives and contaminants. World Health Organization Technical Report Series 710. WHO, Geneva.

WHO. 1997. Zeolites other than Erionite. IARC Monographs on the Evaluation of Carcinogenic Risks to Humans: Silica, some silicates, coal dust and para-aramid fibrils. Vol. 68. World Health Organization, Geneva. pp. 307-327.

Wiechen, A. 1998. Procedure for determining radionuclides in samples of animal feed and animal feed raw materials by gamma spectrometry. F-γ-SPEKT-FUMI-01. Federal Coordinating Office for Soil, Vegetation, Animal Feed and Food of Vegetable or Animal Origin. Federal Ministry for the Environment; Nature Conservation; Building and Nuclear Safety (BUMB). 8 pp.

Wu, Y., Wu, Q., Zhou, Y., Ahmad, H. and Wang, T. 2013a. Effects of clinoptilolite on growth performance and antioxidant status in broilers. Biological Trace Element Research 155(2):228-235.

Wu, Q.J., Wang, L.C., Zhou, Y.M., Zhang, J.F. and Wang, T. 2013b. Effects of clinoptilolite and modified clinoptilolite on the growth performance, intestinal microflora, and gut parameters of broilers. Poultry Science 92(3):684-692.

Wu, Q.J., Zhou, Y.M., Wu, Y.N., Zhang, L.L. and Wang, T. 2013c. The effects of natural and modified clinoptilolite on intestinal barrier function and immune response to LPS in broiler chickens. Veterinary Immunology and Immunopathology 153(1):70-76.

Wu, Q.J., Zhou, Y.M., Wu, Y.N. and Wang, T. 2013d. Intestinal development and function of broiler chickens on diets supplemented with clinoptilolite. Asian-Australasian Journal of Animal Sciences 26(7):987-994.

York, P. 1975. Application of powder failure testing equipment in assessing effect of glidants on flowability of cohesive pharmaceutical powders. Journal of Pharmaceutical Sciences 64 (7):1216-1221.

Zafar, U., Vivacqua, V., Calvert, G., Ghadiri, M. and Cleaver, J.A.S. 2017. A review of bulk powder caking. Powder Technology 313:389-401.

Zarcula, S., Tulcan, C., Samanc, H., Kirovski, D., Cernescu, H. and Mircu, C. 2010. Clinical observations in calves fed colostrum supplemented with clinoptilolite. Lucrări Ştiintifice ŞMedicină Veterinară 53(2):64-69.

7.2 Documentation Not Generally Available

Appendix 2.1_Zeocem_2014_ZeoFeed Clinoptilolite 1g568

Appendix 2.2_Zeocem_2015b_Zeolite Data Sheet

Appendix 2.3_EFSA_2013_Clinoptilolite Flowability

Appendix 2.4_Zeocem_2015c_Manufacturing Process

Appendix 2.5_Internal_External Control Processes

Appendix 2.6a

EL. 2015a. Test Report 15/02461 Clinoptilolite of sedimentary origin - Batch M 181214-1. Ekologické laboratorià, Spisska Nova Ves, Slovakia.

Appendix 2.6b

EL. 2015b. Test Report 15/16295 Clinoptilolite of sedimentary origin - Batch M 300615-1. Ekologické laboratorià, Spisska Nova Ves, Slovakia.

Appendix 2.7a

EL. 2016. Test Report 16/01348 Clinoptilolite of sedimentary origin - Batch M 181215-1. Ekologické laboratorià, Spisska Nova Ves, Slovakia.

Appendix 2.7b

ICT. 2015a. Report of XRD analysis of clinoptilolite in the sample of Clinoptiolite of sedimentary origin -batch M 231214-1. Central Laboratories, Institute of Chemical Technology, Prague.

Appendix 2.8a

ICT. 2015b. Report of XRD analysis of clinoptilolite in the sample of Clinoptiolite of sedimentary origin -batch M 300615-1. Central Laboratories, Institute of Chemical Technology, Prague.

Appendix 2.8b

ICT. 2016. Report of XRD analysis of clinoptilolite in the sample of Clinoptiolite of sedimentary origin - batch M 181215-1. Central Laboratories, Institute of Chemical Technology, Prague.

Appendix 2.9_Zeocem_2016_Clinoptilolite Percent_2001_2015

Appendix 2.10_Zeocem_2016_Levels of Heavy Metals:

As Content in ZeoFeed_2001-2018

Cd Content in ZeoFeed_2001-2018

Hg Content in ZeoFeed_2001-2018

Pb Content in ZeoFeed_2001-2018

Appendix 2.11_Zeocem_2016b_GMP Certificate

Appendix 2.12_Zeocem_2018_Certificate of Feed Registration

Appendix 2.13_Zeocem_2015g_Clinoptilolite 1g568 MSDS

Appendix 2.14_Zeocem_2016c_ZeoFeed Product label

Appendix 3.1_Dioxins

Appendix 3.2_Dioxins_2001-2018

Appendix 3.3_Radionuclides

Appendix 3.4_Constituents of Animal Origin

Submission

CONTINUED

IN

NEXT

Volume

SUBMISSION

CONTINUED

FROM

PREVIOUS

VOLUME

E

Inhaltsverzeichnis | contents

TAB

APPENDIX 2.1 Zeocem - 2014

ZeoFeed Clinoptilolite lg 568

E

ZeoFeed®

Clinoptilolite of sedimentary origin (1g568)

DEFINITION:
An additive for the production of feed mixtures.
Category: "Technological additives",
Function group: g) „Binders",
i) „Anti-caking agents"

COMPOSITION

Clinoptilolite (Hydrated Aluminosilicate) of sedimentary origin

CAS NUMBER: 12173-10-3

MANUFACTURER REGISTRATION NUMBER: SK 100139

DECLARED QUALITATIVE PARAMETER:

Qualitative parameter	Legislative	Validity	Limit value	Praktice Year 2014 ∅ (min., max.)
Content of active materials *				
Clinoptilolite content (%)	Commission Regulation (EC) No 651/2013	Clinoptilolite of sedimentary origin	≥ Min. 80.0	92,0 - 92,0
Dioxin content*				
Amount of dioxins such as PCDD/F (ng WHO- TEQ/kg)	Directive 2002/32/EC as amended	Clinoptilolite of sedimentary origin	Max. 0.75	0,094 (0,094-0,096)
Amount of dioxins such as PCBs (ng WHO-TEQ/kg)	*		*	0,032 (0,031-0,033)
Amount of PCDD/F and PCBs (ng WHO-PCDD/F-PCB-TEQ/kg)	Directive 2002/32/EC as amended	Feed additive from functional group binders, anti-caking agents, and coagulants	Max. 1.5	0,126 (0,125-0,129)
Heavy metal content				
Pb content (mg/kg)	Directive 2002/32/EC as amended	Feed additive from functional group binders, anticaking agents, and coagulants	Max. 30	12,32 (11,37-13,42)
Cd content (mg/kg)	Directive 2002/32/EC as amended	Feed additive from functional group binders, anticaking agents, and coagulants	Max. 2.0	0,0211 (0,0140-0,0259)
As content (mg/kg)	Directive 2002/32/EC as amended	Complete feed	Max. 2.0	1,24 (0,90-1,45)
Hg content (mg/kg)*	Directive 2002/32/EC as amended	Complete feed	Max. 0.1	0,00791 - 0,00560
Mycological evaluation *				
All kinds of blight	Slovak Government regulation Nr. 438/2006	Feed mixture for pig fattening and beef cattle	Max. 150,000 blight spores in 1 g of feed	Negative

ZEOCEM, a. s., 094 34 Bystré 282, Slovak Republic
tel.: +421 57 4492643 • fax: +421 57 4452679
e-mail: zeocem@zeocem.sk • www.zeocem.com

AGRO

E

ZeoFeed®

Clinoptilolite of sedimentary origin (1g568)

Qualitative parameters		Legislative	Validity	Limit value	Reality Year 2014 ∅(min., max.)
Microbiological evaluation *	Salmonella	Slovak Government Regulation Nr. 438/2006	All kinds of feed	0	Negat.
	E.coli	Slovak Governmenr regulation Nr. 438/2006	Feed mixtures		<MDA <1.0. 10^1
Radioactive contamination (Cs^{134} and Cs^{137}) (Bq/kg)		Slovak Government regulation Nr. 438/2006	Feed for pigs, lambs, and other kinds of animals	1,250-5,000	1.3±0.50
Animal proteins *					Negat.
Chemical analysis					
SiO_2 content (%)		PNR 08/13		Min. 62.0	64.6 (62.4- 67.3)
Al_2O_3 content (%)		PNR 08/13		Max. 14.0	12.4 (12.2- 12.5)
Fe_2O_3 content (%)		PNR 08/13		Max. 2.30	1.4 (1.1- 1.6)
CaO content (%)		PNR 08/13		Max. 5.50	3.1 (2.9- 3.1)

* External analysis

Property		ZeoFeed 50		ZeoFeed 200		ZeoFeed 0-0.2 mm		ZeoFeed 0-1mm	
		PNR 08/13	Year 2014 ∅ (min., max.)	PNR 08/13	Year 2014 ∅(min., max.)	PNR 08/13	Year 2014 ∅(min., max.)	PNR 08/13	Year 2014 ∅ (min., max.)
Granularity (oversize) (%)	0.05 mm	Max. 25.0	15.8 (11.8-19.5)						
	0.09 mm			Max. 25.0	13.5 (7.1-18.9)				
	0.1 mm	Max. 5.0	3.38 (2.2-4.7)						
	0.2 mm			Max. 10.0	1.5 (0.5-2.6)	Max. 20.0	4.2 (0.9 – 9.4)	Min. 50.0	63.2 (53.8-73.1)
	0.5 mm								
	1.0 mm							Max. 30.0	11.9 (5.4-20.4)
	2.5 mm								
Ion-exchange capacity (mol/kg)		Min. 0.70	0.83 (0.76-0.92)	Min. 0.70	0.82 (0.75-0.90)	Min. 0.60	0.82 (0.74-0.91)	Min. 0.70	0.82 (0.74-0.91)

Property		ZeoFeed 0.2-0.5 mm		ZeoFeed 0.5-1 mm		ZeoFeed 1-2.5 mm		ZeoFeed 0.2-1 mm	
		PNR 08/13	Year 2014 ∅ (min., max.)	PNR 08/13	Year 2014 ∅ (min., max.)	PNR 08/13	Year 2014 ∅ (min., max.)	PNR 08/13	Year 2014 ∅ (min., max.)
Granularity (oversize) (%)	0.05 mm								
	0.09 mm								
	0.1 mm								
	0.2 mm	Min. 70.0	80.1 (70.4-90.7)					Min. 80.0	91.2 (82.1- 97.5)
	0.5 mm	Max. 25.0	8.9 (4.2-15.8)	Min. 80.0	90.9 (85.1- 96.6)			-	
	1.0 mm			Max. 30.0	19.4 (7.3- 29.3)	Min. 80.0	96.1 (89.5 -99.2)	Max. 5.0	1.9 (0.2-4.6)
	1.25 mm							Max. 0.0	0
	2.5 mm					Max. 25.0	3.8 (0.7 - 9.7)	-	
Ion-exchange capacity (mol/kg)		Min. 0.70	0.82 (0.74-0.91)	Min. 0,70	0.82 (0.74-0.91)	Min. 0,70	0.82 (0.74-0.91)	Min. 0.70	0.82 (0.74-0.91)

AGRO

ZEOCEM, a. s., 094 34 Bystré 282, Slovak Republic
tel.: +421 57 4492643 • fax: +421 57 4452679
e-mail: zeocem@zeocem.sk • www.zeocem.com

E

ZeoFeed®

Clinoptilolite of sedimentary origin (1g568)

Property		ZeoFeed 0,2-1 Plus 0,2-1mm		ZeoFeed 0,5-1 Plus 0,5-1 mm	
		PNR 08/13	Year 2014 ∅ (min., max.)	PNR 08/13	Year 2014 ∅ (min., max.)
Granularity (oversize) (%)	0.05 mm				
	0.09 mm				
	0.1 mm				
	0.2 mm	Min. 70.0	82.7 (70.5- 97.3)		
	0.5 mm			Min. 80.0	95.4 (90.9- 98.8)
	1.0 mm	Max. 30.0	14.9 (4.0- 26.1)	Max. 50.0	35.4 (22.2-48.3)
	1.25 mm				
	2.5 mm				
Ion-exchange capacity (mol/kg)		Min. 0.70	0.82 (0.74-0.91)	Min. 0.70	0.82 (0.74-0.91)

APPLICATION:
ZeoFeed is recommended for all feeds for all animal species.

DOSAGE:
Mix ZeoFeed homogeneously into the feed.
Maximum permissible content 10,000 mg/kg of the complete feed.

STORAGE:
The product must be stored in original, undamaged and closed containers in dry, hygienically clean, well-aired indoor storages, away from foodstuffs, beverages and medicines. Unused ZeoFeed (remains without other additives) can be worked into the soil, since clinoptilolite is a certified soil adjuvant. Clinoptilolite is not a water contaminant - it can also be disposed of in the sewage system. Clinoptilolite does not result in hazardous waste! Any used ZeoFeed packaging or containers, once empty, are to be disposed of as separated waste.

LIFETIME:
If storage conditions are observed, granular ZeoFeed remains usable for up to 2 years.

DELIVERY:
25 kg paper bags (40 pcs / pallet)
BigBag packaging (1,000 kg / 1,200 kg / pallet)
bulk material, silo

ZEOCEM®
ZEOCEM, a. s., 094 34 Bystré 282, Slovak Republic
tel.: +421 57 4492643 • fax: +421 57 4452679
e-mail: zeocem@zeocem.sk • www.zeocem.com

AGRO

E

ZeoFeed®

Clinoptilolite of sedimentary origin (1g568)

CONTACT: Agro-line Project manager
MVDr. Jaroslav Tall
tall@zeocem.sk
tel.: +421 577862 033
fax: +421 574 452 679
mob.: +421 905 903 380

MANUFACTURER: ZEOCEM, a.s., 094 34 Bystré 282, Slovak Republic
Holder of certificate ISO 9001 and FAMI -QS
tel.: 00421/57/4492643, fax: 00421/57/4452679
e-mail: zeocem@zeocem.sk, www.zeocem.com

Version: XF/BXAD

AGRO

Zeolite Data Sheet

Chemical name	Clinoptilolite (Hydrated Aluminosilicate) of sedimentary origin
Empirical formula	$Ca_{1,8} K_{1,8} Na_{0,2} Mg_{0,2} Al_6 Si_{30} O_{72}.24H_2O$ [1] $(Na_{0,21} K_{1,74})(Ca_{1,71} Mg_{0,31})(H_2O)_{18,28} \| [Al_{6,11} Si_{29,90} O_{72}]$ [2] $(Na_{0,08} K_{0,35})(Ca_{0,44} Mg_{0,08})[Al_{1,47} Si_{7,53} O_{18}] . 4,34\ H_2O$ [3]
General formula	$(Ca, K_2, Na_2, Mg)_4 Al_8 Si_{40} O_{96}.24H_2O$

CHEMICAL COMPOSITION

SiO_2	65.00 – 71.30 %		MgO	0.60 – 1.20 %
Al_2O_3	11.50 – 13.10 %		Na_2O	0.20 – 1.30 %
CaO	2.70 – 5.20 %		TiO_2	0.10 – 0.30 %
K_2O	2.20 – 3.40 %			
Fe_2O_3	0.70 – 1.90 %		Si/Al ratio	4.80 – 5.40

MINERAL COMPOSITION

Clinoptilolite	>80 %		Feldspar	±5 %
Clay minerals	<5 %		Other Crystalline Si phases	<10 %

ION EXCHANGEABILITY PROPERTIES

Total exchange	Ca^{2+} 0.64 – 0.98 mol.kg^{-1}	Mg^{2+} 0.06 – 0.19 mol.kg^{-1}
	K^+ 0.22 – 0.45 mol.kg^{-1}	Na^+ 0.01 – 0.19 mol.kg^{-1}
Cation exchange capacity (CEC)		1.20 – 1.50 mol.kg^{-1}

LOSS ON DRYING

< 6%

SELECTIVITY

$Cs^+ > Pb^{2+} > NH_4^+ > Cu^{2+} > Zn^{2+}, Sr^{2+}, Cd^{2+} > Ni^{2+} > Co^{2+}$ [4]

$NH_4^+ > K^+ > Mg^{2+} > Ca^{2+}$ [4]

$Cs^+ > NH_4^+ > Pb^{2+} > K^+ > Na^+ > Ca^{2+} > Mg^{2+} > Ba^{2+} > Cu^{2+} > Zn^{2+}$

PHYSICAL AND MECHANICAL PROPERTIES

Porosity	24 – 32 %	Softing point	1260° C
Effective Diameter of Pores	0.4 nm (4 Å)	Melting point	1340° C
Flow temperature	1420° C	Compactness	70 %
Compressive strength	33 MPa	Whiteness	70 %
Appearance and odour	Grey-green with no odour	Mohs hardness	1.5 – 2.5
Physical state (20° C)	solid	Gridability as per VTI	kVTI = 1.628

DATA ON REACTIVITY

Acid stability	79.50 %	No dangerous decomposition
Thermal stability	up to 400° C	No dangerous polymerisation
Specific surface (BET)	30 – 60 m^2/g	
Solubility in water	none	

[1] Technology of Zeolites production, Final Report, reg. No. K12-526-056

[2] http://www.iza-online.org/natural/Catalog/Slovakia/SlovakiaFrame.htm, Carmine Colella

[3] Nižný Hrabovec – PP Zeolite tuff, Final Report, 1985

[4] Ionenaustausch an Naturzeolithen bei Wasserbehandlungsprozessen, Eva Horváthová-Chmielewská

ZEOCEM®

ZEOCEM, a.s., 094 34 Bystré 282, Slovak Republic
tel.: +421 57 4492643 • fax: +421 57 4452679
e-mail: zeocem@zeocem.sk • www.zeocem.com

ZEOCEM

E

Appendix 2.3. **Effect of clinoptilolite on the flowability of different feedingstuffs**

Type of feed	Feed particle size[a] (X50, mm)	Clinoptilolite (%)	Storage time (Solidification)[b]	Flow factor[c]
Piglet	0.39	0	–	6.4
		1	–	9.7
		2	–	11.5
		0	+	2.2
		1	+	2.9
		2	+	2.8
Piglet	0.46	0	–	4.5
		2	–	4.8
		0	+	1.9
		2	+	2.6
Sow	0.70	0	–	14.0
		2	–	10.7
		0	+	1.7
		2	+	2.1
Cattle	0.58	0	–	36.7
		1	–	14.5
		2	–	50.9
		0	+	2.1
		1	+	2.2
		2	+	2.4
Layer	1.34	0	–	13.0
		2	–	26.8
		0	+	2.3
		2	+	4.0
Dog	0.23	0	–	3.0
		2	–	4.0
Cat	0.19	0	–	4.2
		2	–	5.1
		0	+	2.5
		2	+	2.8

[a] Expressed as the median diameter of 50 % of the particles.
[b] Storage time of one day in a translation shear cell with 1.5 kg load at ambient conditions (18 °C; 60 % relative humidity).
[c] According to Jenike (1967).
Adapted from EFSA (2013).

E

(b) (4)

[redacted]

Elaborated by: Ing. Dzivák, Ing. Mihočová

E

Appendix 2.5 Internal and external process controls

Table 2.5.1 Plan of control system in own laboratory - ZEOCEM

(b) (4)

[redacted]

Table 2.5.1 Plan of control system in own laboratory – ZEOCEM (continued)

(b) (4)

[redacted]

E

Table 2.5.2 Plan of external control - ZeoFeed

(b) (4)

[redacted]

E

(b) (4)

[redacted]

E

(b) (4)

[redacted]

E

(b) (4)

[redacted]

E

(b) (4)

E

(b) (4)

[redacted]

E

(b) (4)

[redacted]

E

(b) (4)

[redacted]

E

(b) (4)

[redacted]

E

(b) (4)

[redacted]

Ap

E

(b) (4)

[redacted]

E

(b) (4)

[redacted]

E

(b) (4)

[redacted]

E

(b) (4)

[redacted]

Appendix 2.9–Zeocem–2016a–Clinoptilolite Percent–2001–2015

E

(b) (4)

[redacted]

E

(b) (4)

[redacted]

E

(b) (4)

[redacted]

E

(b) (4)

[redacted]

E

Certificate NL16/818844200
FAMI-QS Registration Number: FAM-0377

The Feed Safety Management System including Good Manufacturing Practice (GMP) of

Zeocem a.s.

Bystré 282
094 34 Bystré
Slovakia

has been assessed and certified as meeting the requirements of

FAMI-QS Code

(Version 5.1, 14 February 2014)

For :

Technological additives
- binders, anti-caking agents

This certificate is valid from 09/04/2016 until 08/04/2019 and remains valid subject to satisfactory surveillance audits.
Issue 4. Certified since 10/04/2007.
For the validity of this certificate please check www.fami-qs.org

Authorised by

D.J. Verweij
Certification Manager
SGS Product & Process Certification

SGS Nederland B.V.
SGS Product & Process Certification
PO Box 200,
3200 AE Spijkenisse, The Netherlands
t +31(0)181-693297 – f +31(0)181-693572 - www.sgs.com

E

ÚKSÚP

ÚSTREDNÝ KONTROLNÝ A SKÚŠOBNÝ ÚSTAV POĽNOHOSPODÁRSKY V BRATISLAVE

Č.j.: **KVZ/232-003/2018**

V Bratislave, **dňa 15.05. 2018**

OSVEDČENIE

Ústredný kontrolný a skúšobný ústav poľnohospodársky v Bratislave (ďalej len „kontrolný ústav“), ako orgán štátnej správy príslušný na konanie podľa §9 zákona č. 271/2005 Z.z. o výrobe, uvádzaní na trh a používaní krmív (krmivársky zákon) a podľa zákona č. 71/1967 Zb. o správnom konaní (správny poriadok) v znení neskorších predpisov, vydáva v súlade s nariadením Európskeho parlamentu a Rady (ES) č.183/2005, ktorým sa stanovujú požiadavky na hygienu krmív podľa čl.9 a podľa zákona č. 271/2005 Z.z. §4 ods.2, osvedčenie o zápise krmivárskeho podniku do registra.

Registračné číslo **SK 100139**

pre krmivársky podnik **ZEOCEM, a.s.**
IČO: 36 457 728
so sídlom: 094 34 Bystré 282
registrovaná prevádzka: 094 34 Bystré 282

Činnosť:

Kód	Popis
C1V	Registrovaný výrobca kŕmnych surovín
C2V	Registrovaný výrobca kŕmnych doplnkových látok
C3V	Registrovaný výrobca premixov
E	Registrovaný vývozca do tretích krajín

Produkty:

Kategória	Typ
Kŕmne suroviny	11.9.1 – Minerálny grit; Zoolit-universal (register notifikovaných kŕmnych surovín)
Technologické doplnkové látky	1g568 – Klinoptilolit sedimentárneho pôvodu; E 567 – Klinoptilolit sopečného pôvodu
Premixy	Premixy kŕmnych doplnkových látok
Vývoz krmív	1g568 – Klinoptilolit sedimentárneho pôvodu; E 567 – Klinoptilolit sopečného pôvodu; 11.9.1 – Minerálny grit; Zoolit-universal (register notifikovaných kŕmnych surovín); Premixy kŕmnych doplnkových látok

Osvedčenie je platné: **od 27. 09. 2018 do 27. 09. 2023.**

E

Upozornenie:
Podľa zákona č. 271/2005 Z.z., §4 ods.1 krmivársky podnik je povinný každú zmenu údajov zapísaných v registri písomne oznámiť kontrolnému ústavu najneskôr do 30 dní odo dňa, keď ku zmene došlo.

Odôvodnenie:
Na základe žiadosti o registráciu krmivárskeho podniku, ktorú kontrolný ústav prijal a po preskúmaní zistil, že všetky zákonom stanovené náležitosti uvedené v žiadosti boli splnené, vydal krmivárskemu podniku osvedčenie o registrácii. Krmivársky podnik sa vyčiarkne z tohto registra ak vstúpi do platnosti aspoň jeden bod ods.3 §4 zákona č. 271/2005 Z.z.

Poučenie:
Proti tomuto osvedčeniu možno podľa §53 a §54 správneho poriadku podať odvolanie v lehote 15 dní odo dňa jeho doručenia, a to na ÚKSÚP Matúškova 21, 833 16 Bratislava.
Rozhodnutie je preskúmateľné súdom podľa §47 ods.4 správneho poriadku až po využití riadneho opravného prostriedku (odvolania).

ÚSTREDNÝ KONTROLNÝ A SKÚŠOBNÝ ÚSTAV POĽNOHOSPODÁRSKY V BRATISLAVE
SLOVENSKÁ REPUBLIKA
11-1

v.z. [signature]
Ing. Peter Rusňák, PhD.
generálny riaditeľ

Doručuje sa: ZEOCEM, a.s., 094 34 Bystré 282

Osvedčujem, že táto listina doslovne súhlasí s predloženým originálom (~~osvedčeným odpisom~~), pozostávajúcim z -2- strán. Ide o odpis úp... pozostávajúci z -2- strán. Na listine neboli vykonané žiadne zmeny alebo doplnky.
V Prešove dňa 04-06-2013 [4]

JUDr. VLADIMÍR ČUCHTA NOTÁR 7 PREŠOV

JUDr. VLADIMÍR ČUCHTA NOTÁR PREŠOV

UKSUP

CENTRAL CONTROL AND TESTING INSTITUTE
OF AGRICULTURE IN BRATISLAVA

N.: kvz/232-003/2018

in Bratislava, 15/05/2018

CERTIFICATE

The Central Control and Testing Institute of Agriculture in Bratislava (hereinafter as "The Controlling Institute") – Department of feed and Animal Nutrition, as the competent authority pursuant to the § 9 act n. 271/2005 Coll. on production, marketing and utilization of feed ("the act on feed") and pursuant to the act n. 71/1967 coll. on administration proceedings (administration law) as amended by later regulations, issues in accordance with Regulation (EC) n. 183/2005 stipulating the conditions for feed hygiene pursuant to art 9.and pursuant to the act n. 271/2005 coll. sec. 2 it issues this certificate of registration of a feed plant in the register.

Registration number:

SK 100139

For feed production plant: **ZEOCEM, a.s.**
Company n.: 36 457 728
Residing at: 094 34 Bystré 282
Registered plant: 094 34 Bystré 282

Activities:

Code	Description
C1V	Registered feed materials producer
C2V	Registered feed additives producer
C3V	Registered premixtures producer
E	Registered Exporter to the third countries

Products :

Category	Type
Feed material	11.9.1. – Mineral grit Zoolit – universal (register of notified feed materials)
Technologic feed additives	1g568 – Clinoptilolite of sedimentary origin E 567 – Clinoptilolite of volcanic origin
Premixtures	Premixtures for feed additives
Feed exported	1g568 – Clinoptilolite of sedimentary origin E 567 – Clinoptilolite of volcanic origin 11.9.1 – Mineral grit Zoolit – universal (register of notified feed materials) Premixtures of feed additives

The certificate is valid: **from 27/09/2018 to 27/09/2023**

Warning:
Pursuant to the act n. 271/2005 coll. art 4. § 1 the feed plants are obliged to report any changes of data registered in the register in written form within 30 days from the date of change.

Grounds:
According to the request for registration of a feed plant, which was served on the Control Institute, the application was analyzed and the conclusion is that all the feed plant is in conformance with the registration terms and therefore hereby the certificate of registration has been issued to the feed plant. The feed plant will be deleted from the register if at least one point of sec. 3 § 4 of the act n.271/2005 coll.

Instruction:
An appeal can be filed against issuance hereof pursuant to § 53 and § 54 act on administration proceedings within 15 days from serving on UKSUP Matuškova 21, 833 16 Bratislava. This resolution can be reviewed by a court pursuant to § 47 sec. 4 of the administration code after filing the appeal. (remedy)

Round seal print with text:
CENTRAL CONTROL AND TESTING INSTITUTE
OF AGRICULTURE IN BRATISLAVA, SLOVAK REPUBLIC

illegible signature
Ing.Peter Rusňák PhD.
Director general

I certify that this document
Is literally accordant to the presented
original of 2 pages.
The copy is complete of 2 pages.
No correction, alterations, changes
have been made hereto.
In Prešov, date: 04/06/2018

Round seal print with text:
JUDr.VLADIMÍR ČUCHTA, Notary
PREŠOV

Illegible signature

E

Preklad som vypracovala ako prekladateľka zapísaná v zozname znalcov, tlmočníkov a prekladateľov, ktorý vedie Ministerstvo spravodlivosti Slovenskej republiky v odbore slovenský jazyk - anglický jazyk, evidenčné číslo prekladateľa 970399. Preklad je zapísaný pod poradovým číslom 365/18 denníka.
Preklad súhlasí s prekladanou listinou.

V Košiciach, dňa: 04.06. 2018

The Translation has been worked out by the translator registered in the List of the authorised legal experts, interpreters and translators of the Ministry of Justice of the Slovak Republic in the specialisation for Slovak Language - English Language, registration number of the translator 970399. The translation is registered under file number 365/18 of the Journal.
The translation is accordant to the translated document.

In Košice, date: 04. 06. 2018

Mgr. Patricia TROPPOVÁ
PREKLADATEĽ
jazyk: slovenský - anglický

Mgr. Patrícia TROPPOVÁ
PREKLADATEĽ
jazyk:
slovenský - anglický

E

ZEOCEM, a.s. Bystré	Safety Data Sheets
Date of preparation: 16th May 2001	Preparation: **Clinoptilolite of sedimentary origin (1g 568) with trade name ZeoFeed**
Date of revision: 09.06.2004, 05.12.2005, 29.05.2007,15.08.2010,30.11.2010, 19.01.2012. 16.01.2013,21.07.2013, 21.02.2014, 13.02.2015	Page 1 from 8

1. Identification of the substance/mixture and of the company/undertaking

1.1 **Product identifier** (Trade name of the preparation)	Clinoptilolite of sedimentary origin (1g 568) with trade name ZeoFeed
1.2 **Relevant identified uses of the substance or mixture and uses advised against**	ZeoFeed is registered feed additive from: Category: "Technological additives" Functional group: g, "Binders," i, „anticaking agents " Active substance: Clinoptilolite (Hydrated Aluminosilicate) of sedimentary origin Based on the selective adsorption effect, it binds harmful substances (ammonia, $NH4^+$, cadmium, lead and mycotoxines) in the digestive tract of animals. It adsorbs unwished substances of the paunch fermentation, and thus contributes to the pH stabilization in the paunch – buffer effect. It binds unbound water in the feed, and thus decreases the risk of fungus creation. The ammonia load from excrements and urine is reduced in the stable.
1.3 **Details of the supplier of the safety data sheet** (Manufacturer, owner of the preparation registration for protection of stored stocks)	ZEOCEM, a.s. 094 34 Bystré 282, Slovak Republic Tel. No.: 00421 / 57 / 4452414 Fax No.: 00421 / 57 / 4452679 E-mail: zeocem@zeocem.sk
1.4 **Emergency telephone number** Toxicological information centre	Slovak Toxicological Information Centre (Národné toxikologické informačné centrum) University Hospital Bratislava (Univerzitná nemocnica Bratislava), workplace Kramáre Industrial Medicine and Toxicology Clinic (Klinika pracovného lekárstva a toxikológie) Limbová 5, 833 05 Bratislava Tel. No.: +421 2 54652307 Mobile: +421 911 166 066 Fax. No.: +421 2 54774605 e-mail: ntic@ntic.sk

2. Hazards identification

2.1 Classification of the substance or mixture	Product ZeoFeed is produced from natural zeolite of clinoptilolite type. Clinoptilolite (Hydrated Aluminosilicate) of sedimentary origin. It is substance which occur in nature. It is substance according to AnnexV Regulation (EC) No 1907/2006 of the European Parliament and of the Council (exemptions from the obligation to register in accordance with article 2(7)(b).

E

	This substance is not stated in Schedule No. I of Directive No. 67/548/EHS in DSD (Dangerous Substance Directive) and it has not the prescribed classification in compliance with this Directive. This substance is not stated in Table No. 3.1 of Schedule No. VI of Directive ES No. 1272/2008 on classification, labelling and packing of substances and mixtures, and on alteration, amendment and cancellation of Directives No. 67/548/EHS and No. 1999/45/ES and on alteration and amendment of Directive (ES) No. 1907/2006 and it has not any prescribed "harmonized" classification in compliance with this Directive.
2.2 Label elements **according to Directive No. 67/548/EHS DSD**	***ES Classification :*** **This substance has not any prescribed classification according to Directive No. 67/548/EHS DSD** Based on his own decision, the raw material's manufacturer labelled the substance in the following way, as follows: **Warning dangerousness symbol:** it is not given **R-sentences:** it is not given **S-sentences:** S2 Keep it out of range of children. S 36/37 Wear the suitable clothing and gloves.
according to Directive (ES) No. 1272/2008	***GSH classification:*** **This substance has not the prescribed classification according to Directive (ES) No. 1272/2008.** **Based on his own decision, the raw material's manufacturer labelled this substance in the following way, as follows:** **GSH pictograms:** it is not given **Warning word:** it is not given **Warning notice:** it is not given **Safety notice – prevention:** P 102: Keep it out of range of children. P280: Wear the protective gloves / protective clothing / protective glasses. **Safety notice – response:** it is not given **Safety notice – storage:** it is not given **Safety notice – disposal:** it is not given

3. *Composition/information on ingredients*

3.1 Substances	***Trade name: ZeoFeed*** ***Composition: 100 % natural clinoptilolite/*** *Clinoptilolite (Hydrated Aluminosilicate) of sedimentary origin.*
3.2	CAS Number 12173-10-3 Molar mass: not specified

4. First aid measures

If any ailments are manifest, or in case of any doubts, a doctor should be informed about it and information, as per this Safety Data sheet, should be provided with him.

Type of hazard	*Acute danger*	*Prevention*	*First aid*
Aspiration	It is not specified.	By means of the technical measures to ensure that the NPEL of dust does not exceed 5 mg / m^3 in the closed rooms (warehouses). Where it is not possible to ensure temporarily these conditions, it is necessary to use, for protection of respiratory track, the anti-dust respirator.	Fresh air, and take medical advice.
Skin	It does not represent any risk after repeated skin applications and it is not absorb by skin in the harmful quantity.	To use the protective gloves and protective clothing.	To wash the affected place with water and soap.
Eyes	It irritates very mildly and short – lasting the conjunctiva mucosa.	To use the protective glasses.	To wash the eyes with huge amount of water during several minutes, and to take medical advise.
Intake	It is not specified.	Do not eat and drink at the workplace. After work completion, to wash your hands.	To drink 0.5 l of drinking water, or lukewarm water with suspension of the activated charcoal. Take medical advice.

5. *Firefighting measures*

5.1 Extinguishing media	Material is not inflammable. Shall be necessary to be adapted to the substances stored in their close proximity.
5.2 Special hazards arising from the substance or mixture	It is not specified
5.3 Advice for firefighters	It is not specified
Other information	It is not specified

6. *Accidental release measures*

6.1Personal precautions, protective equipment and emergency procedures	Personal preventive actions (safety measures for protection of persons):

E

	To use the personal protective means, and to not breath in any dust.
6.2 Environmental precautions	Environmental precautions: Before its adsorption, the pure Zeolite (without any other mixtures) can be worked into the soil, because Zeolite is the registered soil auxiliary substance. Zeolite does not contaminate even water – it can be also discharged into the sewerage (Zeolite is used as an adsorbent for treatment of drinking water and also for clarification of waste water). Zeolite is not a dangerous waste !
6.3 Methods and material for containment and cleaning up	Mechanically, residues of Zeolite (without any other additives) can be worked into the soil (Zeolite is the certified soil auxiliary substance), or discharged into the sewerage. After its through emptying, the used packs from this preparation should be handed over into the separated collection, or to the approved refuse incinerating plant.

7. *Handling and storage*

7.1 **Precautions for safe handling**	- from the view of safety and health protection in working, the employees must use the personal protective means during manufacturing of this preparation and application thereof, - the working environment, during manufacturing of this preparation and handling with thereof, must be ensured in such a way so that the NPEL of dust has not been exceeded in the air, - the employees must be instructed about product, and they must follow the principles of heath and environment protection.
7.2 **Conditions for safe storage, including any incompatibilities**	This product must be stored in the original and undamaged and closed packs, and in dry, hygienically clean, and in good ventilated and covered warehouses, and separately from foodstuffs. In case of packing for small customers, it should be also stored separately from medicaments and desinfecting agents.

8. *Exposure controls/personal protection*

8.1 Control parameters:

Components

Base	Value	Limiting value	Exposition limiting value
It is not given	It is not given	It is not given	It is not given

8.2 Inspections of exposition – Personal protective means:

In the form of dust (aero-dispersed system) in the working environment, it is necessary to assess Zeolite as a substance with predominantly fibrous effect (respiration limit is 5 μm). NPEL of dust for the working environment is 5 mg.m^{-3} . The workplaces shall be dust exhausted, or ventilated, in such a way so that the maximum dust concentration is 5 mg.m^{-3} of air. Where it is not possible to provide temporarily these conditions, the personal protective working means (protective clothing and shoes, respirator with dust separator, protective

9. Physical and chemical properties

9.1 Information on basic physical and chemical properties	
State at 20 °C	Solid
Colour	Light greyish green

Aroma, malodour	No aroma and malodour
pH	
Boiling temperature	No information is available.
Flash point	Up to 600 °C neg.
Ignition temperature	Settled dust – up to 600 °C neg., raised dust – up to 800 °C neg.
Explosive limits (volume % in the air)	Non-explosive.
Specific weight (kg/m^3)	2200-2440
Oxidation properties	No information is at disposal.
Steam pressure	No information is at disposal.
Solubility	No information is at disposal.
Solubility in water	No information is at disposal.
Distributive coefficient n-octane / water	No information is at disposal.
Viscosity	No information is at disposal.
Vapour density	No information is at disposal.
Evaporation rate	No information is at disposal.
9.2 Other information	
Miscibility	No information is at disposal.
Fat solubility	No information is at disposal.
Conductivity	No information is at disposal.
Melting point	1340 °C
Autoignition temperature	No information is at disposal.

10. Stability and reactivity

10.1 Reactivity	No information is at disposal
10.2**Chemical stability**	No information is at disposal
10.3**Possibility of hazardous reactions**	No information is at disposal
10.4 **Conditions to avoid**	No information is at disposal
10.5**Incompatible materials**	No information is at disposal
10.6 Hazardous decomposition products	No information is at disposal.

11. Toxicological information

Information on toxicological effects	No information is at disposal.
Acute toxicity for substance	
LD_{50}	It was not possible to determine the value of acute oral LD_{50}. After application of dose of 20.000 mg/kg, no animal has died. It is not possible to apply higher doses. It was not possible to determine the value of acute dermal LD_{50}. After application of dose of 5.000 mg/kg on the cut back skin of experimental animals, no animal has died. It is not possible to apply higher doses.
Eye irritability	It irritates very mildly and short-lasting the conjunctiva mucosa. / Apart from the mild congestion after two (2) hours after application of the preparation, which has disappeared within 24 hours, no inflammation changes of conjunctiva mucosa have been observed there /.
Skin irritability	Zeolite does not cause any inflammation changes on the intact

E

	or damaged skin, as well as any other indications of toxicity, even after multiple applications thereof.
Influence on live organisms	Zeolite can be classified as a substance, which is low toxic up to harmless substance. It does not represent any risk even after repeated skin applications, and it does not absorb by skin in the harmful quantity. It irritates very mildly and short – lasting the conjunctive mucosa.
skin corrosion/irritation	Zeolite does not cause any inflammation changes on the intact or damaged skin, as well as any other indications of toxicity, even after multiple applications thereof.
serious eye damage/irritation	It irritates very mildly and short-lasting the conjunctiva mucosa. / Apart from the mild congestion after two (2) hours after application of the preparation, which has disappeared within 24 hours, no inflammation changes of conjunctiva mucosa have been observed there /.
respiratory or skin sensitisation	No information is at disposal.
germ cell mutagenicity	No information is at disposal.
carcinogenicity	No information is at disposal.
reproductive toxicity	No information is at disposal.
STOT-single exposure	No information is at disposal.
STOT-repeated exposure	No information is at disposal.
aspiration hazard	No information is at disposal.
Other Information:	Even other dangerous properties can not be eliminated. It is necessary to handle with this product with such caution like with chemicals.

12. Ecological information

12.1 Toxicity :

The results obtained by testing of Zeolite of Clinoptilolite type did not enable to define the LC 50 for fish and daphnia, because the tested animals have survived the maximum concentrations exceeding the limits for classification of the preparation into the group of substances "for fish and other animals it is almost non-toxic".
Based on the 96-hour static and acute toxicity tests on fish (Cyprinus carpio L., Poecilia reticulata Peters) and 24-hour acute and immobilization test on daphnia (Daphnia magna Straus), the natural Zeolite was classified as to be a substance almost non-toxic for fish and daphnia (Final Report of research No. 53/NRL/T-102).

Pursuant to the expert's opinion of the NRL for pesticides UVM Košice No. 265/2004 NRL/P-1219 rekl., the natural Zeolite is "relatively innocuous for domestic animals, livestock and free living animals" (Z4), "for fish and other water animals, it is almost non-toxic" (Vo4), and "for birds, in case of exceeding the prescribed doses or concentrations, it is relatively harmless" (Vt5). "It is not suitable for earthworm population".
Zeolite of Clinoptilolite type is registered as feed additive (1g568).

12.2 Persistence and degradability	No information is at disposal.
12.3 Bioaccumulative potential	No information is at disposal.
12.4 Mobility in soil	No information is at disposal.
12.5 Results of PBT and vPvB assessment	No information is at disposal.

12.6 Other adverse effects	No information is at disposal.

13. *Disposal considerations*

Waste treatment methods :

The method of waste disposal – mechanically. The residues of Zeolite (without any other additives) can be mixed in the soil, because Zeolite is registered as the soil auxiliary substance. Zeolite does not contaminate even water – it can be also washed away into the sewerage. Zeolite is not a dangerous waste!

14. Information about transport

Sea transport (IMDG): UN No.: UN proper shipping name: Transport hazard class(es): Packing group: Environmental hazards: Special precautions for user: Transport in bulk according to Annex II of MARPOL 73/78 and the IBC Code :	No information is at disposal.
Land transport (ADR): UN No.: UN proper shipping name: Transport hazard class(es): Packing group: Environmental hazards: Special precautions for user: Transport in bulk according to Annex II of MARPOL 73/78 and the IBC Code :	No information is at disposal.
Railway transport (RID): UN No.: UN proper shipping name: Transport hazard class(es): Packing group: Environmental hazards: Special precautions for user: Transport in bulk according to Annex II of MARPOL 73/78 and the IBC Code :	No information is at disposal.
Air transport (ICAO/IATA): UN No.: UN proper shipping name: Transport hazard class(es): Packing group: Environmental hazards: Special precautions for user: Transport in bulk according to Annex II of MARPOL 73/78 and the IBC Code :	No information is at disposal.

15. Regulatory information

Label elements :	

E

according to Directive No. 67/548/EHS DSD	***ES Classification :*** **This substance has not any prescribed classification according to Directive No. 67/548/EHS DSD** Based on his own decision, the raw material's manufacturer labelled the substance in the following way, as follows: **Warning dangerousness symbol:** it is not given **R-sentences:** it is not given **S-sentences:** S2 Keep it out of range of children. S 36/37 Wear the suitable clothing and gloves.
according to Directive (ES) No. 1272/2008	***GSH classification:*** **This substance has not the prescribed classification according to Directive (ES) No. 1272/2008.** **Based on his own decision, the raw material's manufacturer labelled this substance in the following way, as follows:** **GSH pictograms:** it is not given **Warning word:** it is not given **Warning notice:** it is not given **Safety notice – prevention:** P 102: Keep it out of range of children. P280: Wear the protective gloves / protective clothing / protective glasses. **Safety notice – response:** it is not given **Safety notice – storage:** it is not given **Safety notice – disposal:** it is not given
Warning designation	Handling with it is in conformity with good operational hygiene, and principles of safety and health protection in working.

16. Other information

The Security Card contains the data necessary for ensuring the safety and health protection in working and environment protection. The given data corresponds to the present state of knowledge and experience, and they are in conformity with the legal regulations. The particular conditions of using this product at the consumer, however, they are outside of the range of our surveillance and control. The customer is responsible himself/herself for observance of the security provisions.

This Security Data Card has been revised and reworked according to the Regulation of the European Parliament and Council (ES) No. 1907/2006 on registration, evaluation, authorization and limitation of chemical substances (REACH).

E

Clinoptilolite of sedimentary origin (1g568)

Feed Additive

ZeoFeed® 200

Registration number of manufacturer: SK 100139

Category: Technological additives
Functional group: g, Binders
i, Anticaking agents

Active substance (description): Clinoptilolite (Hydrated Aluminosilicate) of sedimentary origin

Fraction: 0-200 μm

Dosing:
ZeoFeed is recommended for all feedingstuffs for all animal species.
ZeoFeed to mix homogenously into the feedingstuffs
Maximum content of ZeoFeed **10 000 mg/kg of complete feedingstuff**

Safety precautions and first aid:
From toxicological point of view can be clinoptilolite classified as a non-toxic substance. It does not represent any hazard following repeated skin applications and is not absorbed through skin in harmful quantities. The worksites have to be de-dusted, or ventilated in such a way that the maximum dust concentration would be 5 $mg.m^{-3}$ of air. At places where it is not possible to secure these conditions temporarily, the should be applied for the protection of respiratory system and skin personal protective working aids /working clothes and shoes, goggles, respirator with anti-dust filler, rubber gloves /. It is necessary to wash hand with soap following the finishing of works.

First aid:
In the case of inhalation should be provided for the afflicted person a stay on fresh air. In the case of accidental consumption you should drink 0,5 l of potable water, or lukewarm water with a suspension of activated carbon. Seek medical aid. In the case of a skin affliction and of clothes take off the clothes and wash the skin by lukewarm water and soap. In the case of an affliction of eyes immediately wash them by a stream of clean water.

Storage
The product should be stored in original undamaged and closed packing, in dry, hygienically clean, well ventilated and closed warehouses, separated from foodstuffs, beverages and medicines.
The not used ZeoFeed (remains without other additives) can be processed into the soil, because clinoptilolite is a certified auxiliary soil substance. Clinoptilolite is not contaminating water – it can be splashed into the sewage system. Clinoptilolite is not a dangerous waste!
The used packing from ZeoFeed, following their thorough emptying, should be handed over to separated collection or should be secured their combustion in a certified incinerator.

In the case of an observance of storage conditions the service life of ZeoFeed is 2 years.

Date of production:.......................................

Batch No:..

Net weight : ....

Version 250913

Producer: ZEOCEM, a. s., 094 34 Bystré 282, SR
Holder of Certificate ISO 9001:2015 and FAMI-QS
phone: 00421/57/4452414. fax: 00421/57/4452679
e-mail: zeocem@zeocem.sk, www.zeocem.sk

For further information contact: G-SCIENCE, Inc., 5500 Highlands Parkway, Smyrna, GA, 30082 USA

(b) (4)

[redacted]

(b) (4)

[redacted]

(b) (4)

[redacted]

E

(b) (4)

[redacted]

E

E

(b) (4)

E

(b) (4)

[redacted]

(b) (4)

[redacted]

E

(b) (4)

[redacted]

(b) (4)

[redacted]

(b) (4)

[redacted]

Submission

<u>CONTINUED</u>

<u>IN</u>

<u>NEXT</u>

Volume

E