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# May the use of Italian volcanic zeolite-rich tuffs as additives in animal diet represent a risk for the human health?

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## Abstract

In Europe the use of zeolite-rich tuffs as additives in animal feeding is well established and ratified by legislation. Quality checks on batches are mandatory to determine the undesirable elements such as lead that, among the heavy metals, plays a primary role for its devastating effects on the life quality of living beings. The present study aimed at determining the total and leached Pb for different samples of Campanian zeolite-rich tuffs related to the most relevant volcanic eruptions (Campanian Ignimbrite and Neapolitan Yellow Tuff). Other samples deriving from other Italian and extra-european exploitation sites were used for comparative tests. The research demonstrated that lead occurrence is linked to the specific paragenesis of the investigated rocks whereas the leached Pb largely depends on the type of zeolite and its amount. It was also established that the use of Campanian zeolite-rich tuffs as additives in animal feeding can be tolerated as these materials are well comparable to analogous products from the minerogenetic point of view such as the clinoptilolite of volcanic origin.

*Key words:* lead; zeolite-rich tuff; undesired elements; animal feeding; Campanian Ignimbrite; Neapolitan Yellow Tuff.

## Introduction

The use of natural zeolites as additives in animal feeding is well established (Mumpton and Fishman, 1977; Pond, 1995; Papaioannou et al., 2005; Colella, 2011). Advantages and disadvantages in the use of 1-6 % clinoptilolite-rich tuffs have been widely discussed in studies carried out directly on animals (Bartko et al., 1995; Yannakopoulos et al., 2000). The main beneficial effects are: I) a growth improvement associated to a rationalization of the feed amounts; II) decrease of gastrointestinal diseases; III) decrease of toxicity towards some heavy metals and ammonium ion; IV) improvement of the quality life in the farm stalls due to the abatement of $NH_4^+$ concentration in the wastes (Mumpton, 1984). Some noxious effects or no significant results were also recorded for some kinds of livestock farming (Chestnut et al., 1992). It should be remarked however, that only in some instances the above mentioned researches faced the problem of the potential toxicity of these additives due to the undesired occurrence of some elements (As, Cd, Hg and Pb), as resulted from the quality check for their use in animal feeding (Mercurio et al., 2009). In particular, these studies refer to the use of additives in swine feeding as binders, anti agglomerants and coagulant agents.

The current use of these additives is reported in the EU regulations which also authorized as zeolitic additives, the Natrolite-Phonolite (E566; European Union Commission Regulation No. 2436-1999), and the Clinoptilolite of volcanic origin (E567; European Union Commission Regulation No. 2148-2004). The first additive is defined as “natural mixture of aluminum silicates, alkalines and alkaline-earth and aluminum hydrosilicates, natrolite (43-46.5 wt.%) and feldspar”; its use is qualified for all the animal species. The second one is described as a “calcium hydrated aluminosilicate of volcanic origin containing minimum 85 wt.% of clinoptilolite and a maximum of 15 wt.% of feldspar, micas and clays, free of fibres and quartz with maximum lead content equal to 80 mg/kg”. In the meanwhile, the European and Italian normative has been updated and new guidelines have been published reporting the law limits allowed for some undesired elements in animal feeding (e.g. maximum leaching Pb equal to 30 mg/kg for E566 and to 60 mg/kg for E567, both values referred to a feeding stuff with a 12% moisture; European Union Commission Directive 2005/87/EC). So far, the two above mentioned mixtures are the only officially authorized zeolite-bearing additives. Nonetheless, other georesources equalized to this category (PHIL-75 equalized to E566; Ministry of Health, 2004) are generally used for this kind of applications such as the zeolite-rich rocks from Campania region, characterized by high phillipsite and chabazite Contents Neapolitan Yellow Tuff (NYT; de’ Gennaro et al., 2000) and Campanian Ignimbrite (CI; Cappelletti et al., 2003). As these rocks are of volcanic origin they bear quite high lead contents thus representing a potential source of health risk. On this account, the present research has a double objective: I) to verify the analytical procedures useful to evaluate the leached Pb as scheduled in the law or methods recognized in Europe (UNI, 2008; Semeraro et al., 2009) considering that the analytical result could be affected by the solid-to-liquid ratio and by the grain size; II) to evaluate the possible use of other zeolite-rich tuffs, further than those already commercialized (i.e. clinoptilolite-rich materials) in this sector of animal diet, through a comparison of their Pb content (either total or leachable).

A further spin-off of the present research, namely the recognition of the risk in using natural zeolites, can be found in the: I) biomedical sector, where zeolites have been proposed for the development of substrates for antibacterial and antidiarrhoic carriers, or slow release drugs in the direct treatment of some pathologies (Colella, 2011), and in the II) oenological sector, where natural zeolites were successfully proposed for optimization of tartaric and protein stability, improving white wines quality (Mercurio et al., 2010).

## Experimental

### *Materials*

Sample collection was carried out bearing in mind the wide availability of materials within the Campania region. As far as the evaluation methods of Pb leachable from the materials usually used in zootechny are concerned, it should be remarked that the factors most affecting the measurements are the grain size of the additive and the solid-to-liquid ratio. On this account, eight grain sizes of a commonly marketed zeolite-rich tuff (Campanian Ignimbrite from *Comiziano*, *Nola* - Naples) have been used (∅ (mm) < 5, AM; 200 < ∅ < 500, BM; 500 < ∅ < 1000, CM; 1000 < ∅ < 1400, DM; 1400 < ∅ < 2000, EM; 2000 <∅ < 3150, FM; 3150 < ∅ < 4000, GM; ∅ > 4000, HM). The same tests were also carried out, as comparative check, on 23 samples collected in different outcrops of CI and on 3 samples of NYT, scattered over the whole Campanian territory.

As further comparative samples, zeolite-rich tuffs from other Italian regions (i.e. Umbria and Lazio) linked to the *Tufo Giallo della Via Tiberina* (TGVT) eruption and the *Ignimbrite di Orvieto Bagnoregio* (IOB) products (Cappelletti et al., 1999) were used. Moreover a clinoptilolite-rich tuff from an exploitation site of Turkey widely commercialized in Italy by *Italiana Zeoliti* company as IZCLINO was characterized (de Gennaro et al., 2008).

Reference pure samples representative of the Campanian zeolite-rich tuffs were obtained by enrichment processes. Starting from a phillipsite-bearing rock (LS0), and a chabazite-bearing one (LS8) the beneficiation was achieved by means of a pilot plant scale mineral processing following the procedure described by de' Gennaro and Franco (1979), paying attention however at avoiding the use of heavy liquids actually replaced by a separation method based on different settling gravity (Chipera et al., 1993). The reference pure sanidine (ORT) was obtained by a stereo microscope hand picking separation on a grey facies CI sample.

### *Lead leaching and digestion tests*

Total Pb by digestion: in Teflon tubes about 0.1 g of powder were first reacted with 25 mL of HF (Carlo Erba Reagents, assay 38-40%, CAS [7664-39-3]) and then with 5 mL of $HClO_4$ (JT Baker Reagents, assay 70-72%, CAS [7601-90-3]). The resulting solution was placed on a sand bath for about 5 hours at 70 °C. After the addition of 20 mL of HF the sample was dried. Finally, once cooled, the solution was added with about 10 mL of sub-boiled distilled water and 2 mL of HCl (Carlo Erba Reagents, assay 35-37%, CAS [7647-01-0]), placed on a sand bath until the dry sample was dissolved and a final 50 mL volume solution was achieved.

Leached Pb: the test was carried out using 5% w/w solutions of $HNO_3$ (Romil-UPA, assay 67-69%, CAS [7697-37-2]) and brought on the boil for 30 minutes; seven different mass samples (1 to 7 g, 1 g steps) have been used. This leaching test was set up according to norm UNI EN 15510: 2008, point 9.1.4. (see UNI EN norm for further details).

Both the above described methods (Total Pb by digestion and Leached Pb) enable to evaluate a factor defined as "available leached Pb" ($ALPb_{meas}$ = Leached Pb/ Total Pb) which gives an overall attitude of the sample to release lead, according to the above described methods.

### *Analytical methods*

Qualitative and quantitative evaluation of the mineralogical components was carried out by X-Ray Powder Diffraction (XRPD) using Reference Intensity Ratio (RIR) and Rietveld methods (Chipera and Bish, 1993; Chipera and Bish, 1995) on a Panalytical X'Pert Pro MPD equipped with a RTMS X'Celerator detector (CuKα radiation, 40 kV, 40 mA, 2Θ range from 3 to 80°, equivalent step size: 0.0179 2Θ, counting time: 120 s per step). Powders with grain size <10 μm were obtained by means of a McCrone micronising mill (agate cylinders and wet grinding time 15 min). An $\alpha$-$Al_2O_3$ internal standard (1 μm, Buehler Micropolish) was added to each sample in amount of 20 wt.%. Inductively Coupled Plasma - Optical Emission Spectroscopy (ICP-OES) analyses allowed to measure total and leached Pb using a Perkin Elmer Optima 2100 Dual View equipped with quartz cyclonic chamber operated in standard mode. Operating parameters are given in Table 1.

The implementation of the calibrating curve was carried out with a 100 ppb lead standard by Perkin Elmer whereas the instrument calibration was performed by reading four solutions with known concentrations (10, 50, 100 and 200 ppb).

The above described procedures account for an uncertainty of 15% for leaching tests, following the method described in Semeraro et al. (2009) and of 5% for digestion tests, according to the instrumental and methodological features.

## Results and discussion

### *Evaluation of check quality procedures*

Table 2 and Figure 1 show the results of the mineralogical analysis and total Pb contents measured on the different commercial grain size fractions, as provided by the manufacturer. Total lead is on average 60 mg/kg, ranging between 53 and 67 mg/kg. Total zeolite (phillipsite + chabazite + analcime) ranges from 60 wt.% for the FM and CM samples to 70 wt.% for the EM sample. The above reported Pb values, in good agreement with literature data (Civetta et al., 1997), evidence relationship neither with the grain size fraction nor with the zeolite content (Figure 1).

Conversely, the results of the leaching tests support the close relationship occurring between the leached lead content, the size fraction, and the solid-to-liquid ratio. In fact, Figure 2, reporting leached Pb as a function of the solid-to-liquid ratio and of the grain size fraction, allows to draw the following considerations: i) regardless the grain size fraction, the amount of leached Pb per unit mass of tested material progressively decreases as the total mass of treated powder increases and the $HNO_3$ volume is unchanged, according to norm (UNI, 2008); ii) at a parity of powder mass, the highest and lowest values of leached Pb always refer to the smallest (AM= micronized; ∅<5) and the largest (HM= ∅> 4000) grain size, respectively. The

Table 1. ICP-OES operating parameters for Pb evaluation.

**Perkin Elmer Optima 2100 Dual View**

| Parameter | Value | Parameter | Value |
|---|---|---|---|
| Purge gas flow | Normal | Plasma flow (argon) L/min | 15 |
| Read Delay Time (sec) | 60 | Aux flow (nitrogen) L/min | 0.2 |
| Replicates | 3 | Nebulizer flow L/min | 0.8 |
| Pb wavelength (nm) | 220.353 | Power watts | 1300 |
| Source equilibration delay (sec) | 15 | Plasma View | axial |
| Plasma aerosol type | wet | Sample flow rate | 1.5 mL/min |
| Nebulizer start-up conditions | instant | Sample flush time (sec) | 15 |

Table 2. XRPD quantitative mineralogical analyses and total Pb values for the eight grain size fractions from Comiziano (Naples).

| ID Sample | Size Fraction (μm) | Total Pb (mg/kg) | Zeolite content (wt.%) | sm (wt.%) | bt (wt.%) | san (wt.%) | phi (wt.%) | cha (wt.%) | ana (wt.%) | Total (wt.%) | am (wt.%) |
|---|---|---|---|---|---|---|---|---|---|---|---|
| AM | ∅ < 5 | 53.0 | 68 | | tr | 28 (±6) | 39 (±2) | 28 (±1) | 1 (±1) | 96 (±9) | 4 |
| BM | 200 < ∅ < 500 | 58.0 | 63 | | tr | 32 (±6) | 35 (±2) | 27 (±1) | 1 (±1) | 95 (±5) | 5 |
| CM | 500 < ∅ < 1000 | 55.5 | 60 | | tr | 34 (±5) | 35 (±2) | 24 (±1) | 1 (±1) | 94 (±9) | 6 |
| DM | 1000 < ∅ < 1400 | 62.0 | 61 | | tr | 26 (±5) | 38 (±2) | 21 (±1) | 2 (±1) | 87 (±8) | 13 |
| EM | 1400 < ∅ < 2000 | 67.0 | 70 | | tr | 25 (±5) | 31 (±2) | 39 (±2) | | 95 (±5) | 5 |
| FM | 2000 < ∅ < 3150 | 58.5 | 60 | | tr | 39 (±8) | 44 (±2) | 15 (±1) | 1 (±1) | 99 (±11) | 1 |
| GM | 3150 < ∅ < 4000 | 66.0 | 62 | 1 (±1) | tr | 28 (±6) | 38 (±2) | 23 (±1) | 1 (±1) | 92 (±9) | 8 |
| HM | ∅ > 4000 | 60.0 | 64 | 1 (±1) | tr | 30 (±6) | 37 (±2) | 26 (±1) | 1 (±1) | 96 (±10) | 4 |

sm = smectite; bt = biotite; san = sanidine; phi = phillipsite; cha = chabazite; ana = analcime; am = amorphous

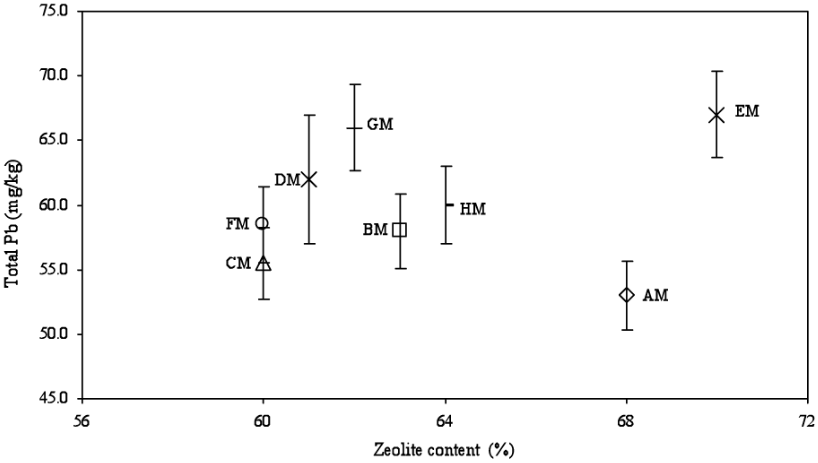


Figure 1. Zeolite content vs. Total Pb for the eight grain size fractions. Labels as in Table 1. Error bar = ± 5%.

2005/87/EC.

*Validation of available and leachable lead in zeolite-rich tuffs*

Table 3 reports the mineralogical composition of the selected samples, mostly belonging to the Campanian volcaniclastic formations of the Campanian Ignimbrite (CI) and the Neapolitan Yellow Tuff (NYT); samples from similar deposits from Latium and Umbria regions, were used as comparative samples along with a described $ALPb_{meas}$ parameter (3c). The examination of Figure 3a enables to state for the Campanian tuffs that: I) the highest total Pb values are recorded in feldspar-bearing tuffs and that Pb increases with sanidine content; II) phillipsite- and chabazite-bearing materials show quite comparable but almost variable total Pb values (max 47 mg/kg ETEP 14; min 22.8 mg/kg COMIZIANO 3) and these values do not correlate with the total zeolite content; III) samples with dominant amorphous matter showed Pb values of

Table 3. Geological and geographical settings along with quantitative mineralogical analyses of the entire set of investigated samples.

| ID Sample | Provenance | Formation | Location | sm (wt.%) | bt (wt.%) | san (wt.%) | phi (wt.%) | cha (wt.%) | ana (wt.%) | cli (wt.%) | Total | am (wt.%) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| SOR 6* | Italy, Campania | CI | Sordina (SA) | | | 14 (±2) | | | | | 14 (±2) | 86 |
| SOR 12* | Italy, Campania | CI | Sordina (SA) | | 2 (±1) | 93 (±14) | | | | | 95 (±14) | 5 |
| PUC 12* | Italy, Campania | CI | Pucara (SA) | | tr | 98 (±13) | | | | | 98 (±13) | 2 |
| SA 3* | Italy, Campania | CI | San Anna (SA) | | | 88 (±11) | | | | | 88 (±11) | 12 |
| DUG 5* | Italy, Campania | CI | Dugenta (BN) | | | 11 (±2) | | | 1 (±1) | | 12 (±2) | 88 |
| ETEP 13 | Italy, Campania | CI | Dugenta (BN) | 4 (±1) | | 24 (±5) | 10 (±1) | 57 (±3) | 1 (±1) | | 96 (±9) | 4 |
| ETEP 14 | Italy, Campania | CI | Dugenta (BN) | | | 25 (±5) | 11 (±1) | 46 (±3) | 1 (±1) | | 83 (±8) | 17 |
| SM 6* | Italy, Campania | CI | S. Mango (AV) | 2 (±1) | 1 (±1) | 17 (±2) | 8 (±1) | 69 (±4) | | | 96 (±5) | 4 |
| PPU 3* | Italy, Campania | CI | Prata di P. U. (AV) | 7 (±1) | 2 (±1) | 17 (±3) | 7 (±1) | 69 (±3) | | | 102 (±5) | |
| BAL 0B* | Italy, Campania | CI | Balzarama (CE) | | 1 (±1) | 37 (±6) | 19 (±1) | | 4 (±1) | | 61 (±6) | 39 |
| BAL 2B* | Italy, Campania | CI | Balzarama (CE) | 5 (±1) | 1 (±1) | 30 (±5) | 34 (±1) | 10 (±1) | 10 (±1) | | 90 (±5) | 10 |
| BAL 5B* | Italy, Campania | CI | Balzarama (CE) | 5 (±1) | 1 (±1) | 26 (±4) | 32 (±1) | 11 (±1) | 7 (±1) | | 82 (±4) | 18 |
| BAL 10B* | Italy, Campania | CI | Balzarama (CE) | 6 (±1) | 1 (±1) | 22 (±3) | 18 (±1) | 41 (±2) | 1 (±1) | | 89 (±4) | 11 |
| AV 3* | Italy, Campania | CI | Aversa (CE) | 9 (±2) | 6 (±1) | 31 (±4) | 29 (±1) | 24 (±1) | | | 99 (±5) | 1 |
| AV 7* | Italy, Campania | CI | Aversa (CE) | | 2 (±1) | 86 (±11) | | | | | 88 (±11) | 12 |
| AT 6* | Italy, Campania | CI | Comiziano (NA) | 6 (±1) | 1 (±1) | 28 (±4) | 42 (±1) | 24 (±1) | 2 (±1) | | 103 (±5) | |
| SZ 1 | Italy, Campania | CI | Comiziano (NA) | 4 (±1) | tr | 28 (±5) | 38 (±2) | 16 (±1) | 2 (±1) | | 88 (±9) | 12 |
| SZ 2 | Italy, Campania | CI | Comiziano (NA) | | tr | 30 (±6) | 38 (±2) | 15 (±1) | 1 (±1) | | 84 (±9) | 16 |
| COMIZIANO 1 | Italy, Campania | CI | Comiziano (NA) | | tr | 31 (±6) | 31 (±2) | 19 (±1) | 1 (±1) | | 82 (±9) | 18 |
| COMIZIANO 2 | Italy, Campania | CI | Comiziano (NA) | | tr | 20 (±4) | 46 (±2) | 10 (±1) | | | 76 (±7) | 24 |
| COMIZIANO 3 | Italy, Campania | CI | Comiziano (NA) | | tr | 26 (±5) | 31 (±2) | 22 (±1) | 5 (±1) | | 84 (±8) | 16 |
| COMIZIANO 4 | Italy, Campania | CI | Comiziano (NA) | | tr | 27 (±5) | 30 (±1) | 28 (±2) | 4 (±1) | | 89 (±8) | 11 |
| COMIZIANO 5 | Italy, Campania | CI | Comiziano (NA) | | tr | 32 (±6) | 17 (±1) | 5 (±1) | 2 (±1) | | 56 (±8) | 44 |
| CAVA V | Italy, Campania | NYT | Quarto (NA) | | 2 (±1) | 19 (±2) | 45 (±1) | 10 (±1) | | | 76 (±3) | 24 |
| CAVA N | Italy, Campania | NYT | Quarto (NA) | | 2 (±1) | 45 (±3) | 10 (±1) | 6 (±1) | 22 (±1) | | 85 (±4) | 13 |
| SAV 3 | Italy, Campania | NYT | Cantarelle (NA) | 5 (±3) | tr | 18 (±4) | 55 (±2) | 1 (±1) | 13 (±2) | | 92 (±5) | 9 |
| C4_1 | Italy, Latium | TGVT | Riano (Roma) | 2 (±1) | | 10 (±1) | 2 (±1) | 49 (±3) | | | 75 (±5)[a] | 25 |
| C2_P1_2 | Italy, Latium | TGVT | Riano (Roma) | 18 (±3) | | 8 (±3) | 10 (±1) | 43 (±2) | | | 92 (±8)[a] | 8 |
| C1_P1_1 | Italy, Latium | IOB | Lubriano (VT) | | 1 (±1) | 25 (±6) | 6 (±1) | 26 (±2) | 5 (±1) | | 73 (±8)[b] | 27 |
| Aff_1_1 | Italy, Umbria | IOB | Bardano (TR) | | 1 (±1) | 25 (±13) | 1 (±1) | 22 (±5) | 3 (±1) | | 60 (±11)[c] | 40 |
| IZ CLINO | Turkey | | Eskisehir | 4 (±1) | tr | 16 (±2) | | | | 59 (±3) | 93 (±8)[d] | 7 |

sm = smectite; bt = biotite; san = sanidine; phi = phillipsite; cha = chabazite; ana = analcime; cli = clinoptilolite; am = amorphous.
[a]calcite; [b]analcime, pyroxene; [c]analcime, pyroxene, amphibole; [d]quartz, opal. *(from Langella et al., 2012).

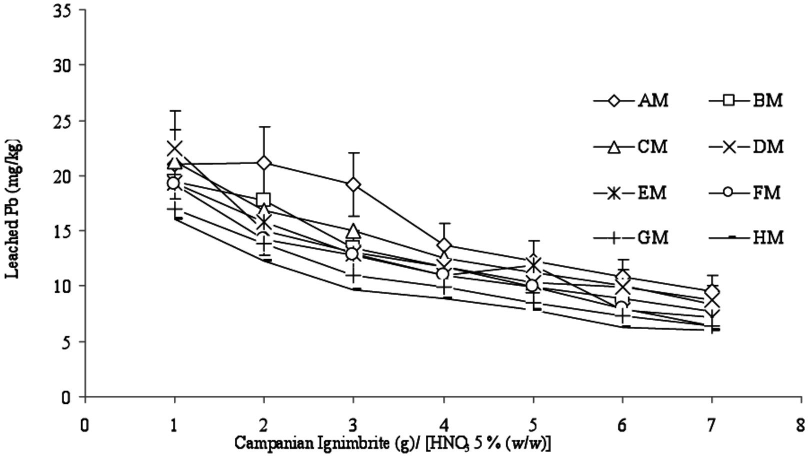


Figure 2. Leached Pb as a function of the solid-to-liquid ratio and of the grain size fraction (labels as in Table 2). Error bar = ± 15% reported only for the highest lead values.

26 mg/kg (SOR6) and 32 mg/kg (DUG5); IV) samples without a clear dominant phase (BAL0B, BAL2B, CAVA V, CAVA N, COMIZIANO 5) gave quite variable Pb values, the lowest in samples from the CI (22.63 mg/kg COMIZIANO 5) the highest for those ascribed to the NYT (58.90 mg/kg CAVA N).

As far as the comparative Italian samples are considered (C4_1, C2_P1_2, C1_P1_1, Aff_1_1) their total Pb contents are however the highest compared to the Campanian samples (CI and NYT). Finally, the clinoptilolite sample (IZCLINO) gave values of about 50 mg/kg.

Leaching tests demonstrate that sanidine, although recording the highest total Pb content, is the phase that releases the lowest amount of this metal (Figure 3b). The amorphous phase also displays a similar behavior although the starting total Pb content is lower. Materials that have phillipsite + chabazite content higher than 50% show a strongly variable behavior but, in any case providing the highest values of leached Pb (Figure 3b).

Relationships between the dominant phase and $ALPb_{meas}$ are well depicted in the diagram of Figure 3c where three distinct areas (I, II, III) can be evidenced. Fields I and II, characterized by a very narrow range of $ALPb_{meas}$ values, are referred to sanidine- or amorphous- dominant samples whereas a third one (III) gathers all the zeolite-rich tuffs samples and can be distinguished for a quite large variability.

Table 4 and Figures 3 (a, b, c) also reports three samples that should be considered as pure phase references (chabazite-rich: LS8 (~95 wt.%); phillipsite-rich: LS0 (~98 wt.%); Sanidine: ORT (100 wt.%)); the $ALPb_{meas}$ values for these reference samples were 0.70 (LS8), 0.95 (LS0) and 0.05 (ORT) and were used for the elaboration of the $ALPb_{calc}$ parameter as later described.

Starting from the $ALPb_{meas}$ values ($ALPb_{meas}$ = Leached Pb/Total Pb; Table 4) measured for each group of dominant phase, the mean values ($ALPb_{av}$) were calculated; they vary between $ALPb_{av}$=0.05 for the dominant feldspar group (this value was perfectly converging with the one measured for the reference material ORT) and $ALPb_{av}$=0.52 for the dominant phillipsite group. Slight lower values were measured for the

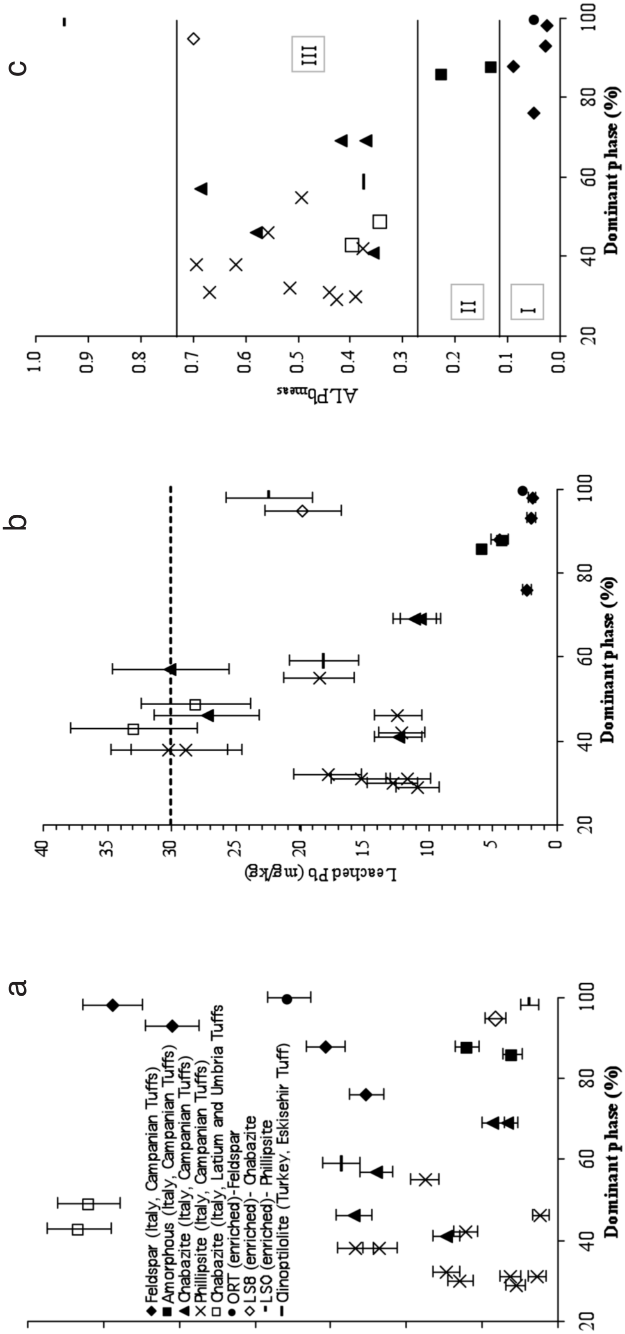
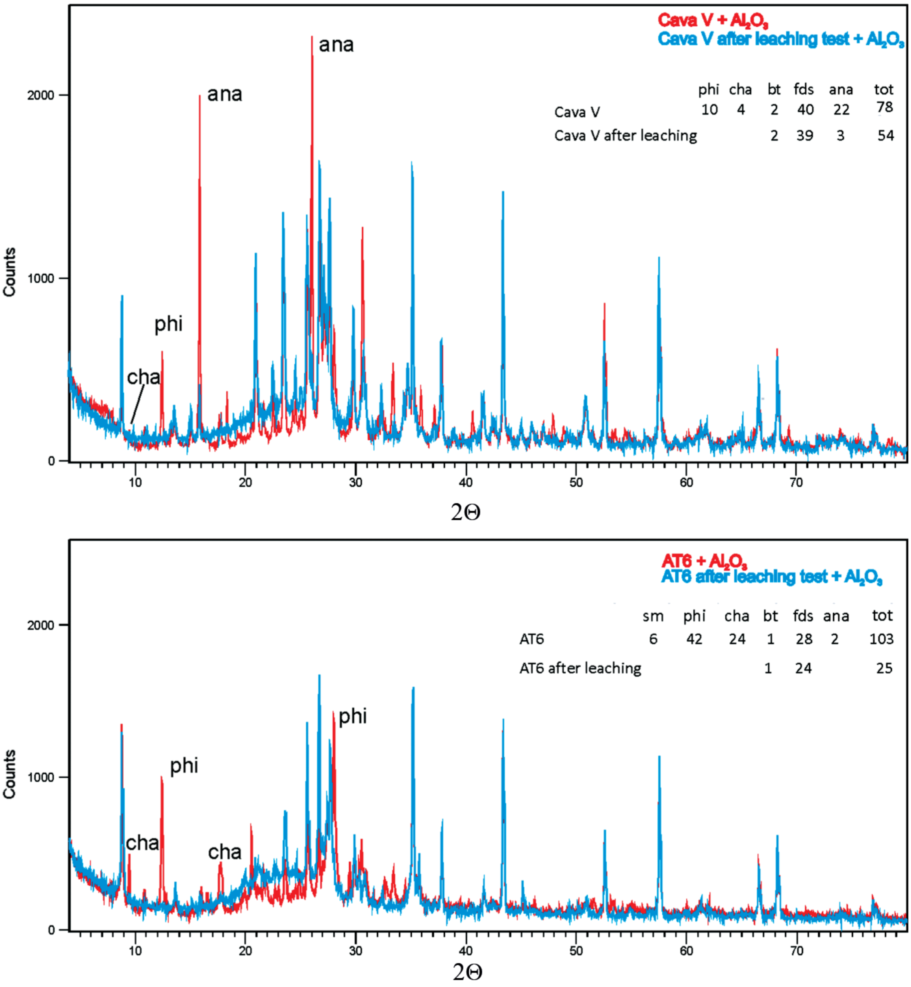


Figure 3. a) Dominant phase vs. Total Pb Error bar = ± 5%; b) Dominant phase vs. Leached Pb. Error bar = ± 15%; c) Dominant phase vs. ALPb$_{meas}$.

Table 4. Leached and total Pb along with ALPb parameters reported for investigated samples grouped as a function of the dominant phase. Reference pure phases (ORT, LS8, LS0) are also reported.

| ID Sample | dominant phase | am (%) | phi (%) | cha (%) | cli (%) | san (%) | Leached Pb (mg/Kg) | Total Pb (mg/Kg) | $ALPb_{meas}$ | $ALPb_{av}$ | $ALPb_{calc}$ | εALPb (%) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| PUC 12 | sanidine | 2 | | | | **98** | 1.94 | 78.76 | 0.02 | | 0.05 | -2.80 |
| SOR 12 | sanidine | 5 | | | | **93** | 2.00 | 71.00 | 0.03 | | 0.06 | -2.73 |
| SA3 | sanidine | 12 | | | | **88** | 4.51 | 50.68 | 0.09 | 0.05 | 0.07 | 2.34 |
| AV7 | sanidine | 12 | | | | **76** | 2.31 | 45.30 | 0.05 | | 0.06 | -0.86 |
| *ORT (enriched)* | *sanidine* | | | | | ***100*** | *2.60* | *55.50* | *0.05* | | *0.05* | -0.32 |
| DUG 5 | amorphous | **88** | | | | 11 | 4.22 | 32.00 | 0.13 | 0.18 | 0.16 | -3.20 |
| SOR 6 | amorphous | **86** | 1 | | | 14 | 5.84 | 26.00 | 0.22 | | 0.17 | 5.33 |
| SM6 | zeolite (chabazite) | 4 | 8 | **69** | | 17 | 11.09 | 26.60 | 0.42 | 0.48 | 0.57 | -15.78 |
| PPU 3 | zeolite (chabazite) | | 8 | **69** | | 17 | 10.63 | 28.60 | 0.37 | | 0.57 | -19.58 |
| ETEP 13 | zeolite (chabazite) | 4 | 10 | **57** | | 24 | 30.10 | 44.00 | 0.68 | | 0.51 | 17.09 |
| ETEP 14 | zeolite (chabazite) | 17 | 11 | **46** | | 25 | 27.27 | 47.00 | 0.58 | | 0.47 | 11.06 |
| BAL10B | zeolite (chabazite) | 11 | 18 | **41** | | 22 | 12.34 | 34.70 | 0.36 | | 0.49 | -13.32 |
| *LS8 (enriched)* | *chabazite* | *5* | | *95* | | | *19.80* | *28.30* | *0.70* | | *0.67* | 2.56 |
| SAV 3 | zeolite (phillipsite) | 9 | **55** | 1 | | 18 | 18.54 | 37.54 | 0.49 | 0.52 | 0.55 | -6.08 |
| COMIZIANO 2 | zeolite (phillipsite) | 24 | **46** | 10 | | 20 | 12.40 | 22.30 | 0.56 | | 0.56 | -0.41 |
| AT 6 | zeolite (phillipsite) | | **42** | 24 | | 28 | 12.13 | 32.20 | 0.38 | | 0.58 | -20.43 |
| SZ1 | zeolite (phillipsite) | 12 | **38** | 16 | | 28 | 28.88 | 46.70 | 0.62 | | 0.51 | 10.98 |
| SZ2 | zeolite (phillipsite) | 16 | **38** | 15 | | 30 | 30.21 | 43.50 | 0.69 | | 0.51 | 18.47 |
| BAL5B | zeolite (phillipsite) | 18 | **32** | 11 | | 26 | 17.87 | 34.70 | 0.51 | | 0.43 | 8.86 |
| COMIZIANO 1 | zeolite (phillipsite) | 18 | **31** | 19 | | 31 | 11.60 | 26.32 | 0.44 | | 0.48 | -3.47 |
| COMIZIANO 3 | zeolite (phillipsite) | 16 | **31** | 22 | | 26 | 15.27 | 22.80 | 0.67 | | 0.49 | 17.94 |
| COMIZIANO 4 | zeolite (phillipsite) | 11 | **30** | 28 | | 27 | 12.82 | 32.95 | 0.39 | | 0.51 | -12.52 |
| AV3 | zeolite (phillipsite) | 1 | **29** | 24 | | 31 | 10.87 | 25.58 | 0.42 | | 0.46 | -3.59 |
| *LS0 (enriched)* | *phillipsite* | *2* | ***98*** | | | | *22.40* | *23.70* | *0.95* | | *0.93* | 1.05 |
| COMIZIANO 5 | no dominant phase | 44 | 17 | 5 | | 32 | 11.40 | 22.63 | 0.50 | | 0.29 | 21.21 |
| BAL0B | no dominant phase | 39 | 19 | | | 37 | 10.55 | 26.38 | 0.40 | | 0.27 | 13.07 |
| BAL2B | no dominant phase | 10 | 34 | 10 | | 30 | 14.49 | 28.45 | 0.51 | | 0.43 | 8.33 |
| CAVA N | no dominant phase | 13 | 10 | 6 | | 45 | 29.94 | 58.90 | 0.51 | | 0.18 | 32.54 |
| CAVA V | no dominant phase | 24 | 45 | 10 | | 19 | 22.30 | 55.40 | 0.40 | | 0.55 | -14.77 |
| C4_1 | chabazite | 25 | 2 | **49** | | 10 | 28.14 | 82.00 | 0.34 | 0.37 | | |
| C2_P1_2 | chabazite | 8 | 10 | **43** | | 8 | 32.98 | 83.23 | 0.40 | | | |
| C1_P1_1 | no dominant phase | 27 | 6 | 26 | | 25 | 23.85 | 80.68 | 0.30 | | | |
| Aff_1_1 | no dominant phase | 40 | 1 | 22 | | 25 | 41.08 | 105.00 | 0.39 | | | |
| IZCLINO | clinoptilolite | 7 | | | **59** | 16 | 18.13 | 48.57 | 0.37 | | | |



| | phi | cha | bt | fds | ana | tot |
|---|---|---|---|---|---|---|
| Cava V | 10 | 4 | 2 | 40 | 22 | 78 |
| Cava V after leaching | | | 2 | 39 | 3 | 54 |

| | sm | phi | cha | bt | fds | ana | tot |
|---|---|---|---|---|---|---|---|
| AT6 | 6 | 42 | 24 | 1 | 28 | 2 | 103 |
| AT6 after leaching | | | | 1 | 24 | | 25 |

Figure 4. XRPD patterns of CAVA V (up) and AT6 (bottom) samples before and after the leaching tests

dominant chabazite group ($ALPb_{av}$=0.48) followed by the dominant amorphous group ($ALPb_{av}$=0.18). Comparative samples provided intermediate values ($ALPb_{av}$=0.37 for both chabazite- or clinoptilolite-bearing rocks).

The above reported considerations lead to infer that zeolite-rich tuffs (mainly the phillipsite-prevailing ones), during the leaching tests release the maximum Pb amounts, although they do not show the highest total Pb contents. The attribution of this behaviour to exchange phenomena occurring during the leaching process do not appear possible. Actually literature data report that both phillipsite (Pansini et al. 1996) and chabazite (Colella and Pansini, 1988; Pansini and Colella, 1990; Torracca et al., 1998) exhibit a large affinity towards $Pb^{2+}$, which makes extremely unlikely its exchange for hydroxonium ions during the leaching test. Moreover, the hypothesis of the occurrence of such exchange was not confirmed by the exchange tests carried out on the same zeolite-rich tuffs using the methodology currently used by this research group (Cerri et al., 2002) which gave back values of exchanged Pb close to the instrumental and methodological detection limits (L.O.D. > 0.1 ppm for AAS). This aspect led us to consider the Pb release as due to other factors such as a loss of crystal structure as a consequence of the acid environment set up during the leaching test (UNI, 2008). On this account, XRPD analyses (Figure 4) carried out on the powders recovered after the leaching tests (CAVA V and AT 6) clearly demonstrate the total breakdown of the zeolite phases, with the only exception of analcime still recorded in very low amount (about 3%, sample CAVA V). It should be remarked however, that this phase only partially contributes to the possible exchange process. The Pb previously wrongly defined as leached element, should be therefore considered as a consequence of the total zeolite framework breakdown thus simulating a total lead obtained by digestion.

The above considerations would lead to state that the investigated materials, cannot be used as additive to animal feeding as they show quite high Pb contents, mainly due to their volcanic origin. The distribution of this element varies among the different occurring phases, being particularly concentrated in sanidine and in the amorphous matter, subordinately. Moreover, the total Pb of samples characterized by the highest amorphous component is almost similar to that measured in high zeolite grade samples, thus confirming the origin of these silicates by transformation (hydrolysis, dissolution) of the glassy matrix (Langella et al., 2012). More difficult to explain is the lack of correlation between the zeolite content and the leached Pb, even more evident when rocks show a more complex paragenesis, without any dominant phase.

An attempt to determine the contribution of each single component of a poliphasic mixture to the leached Pb was carried out by elaborating the $ALPb_{calc}$ parameter as hereafter reported,

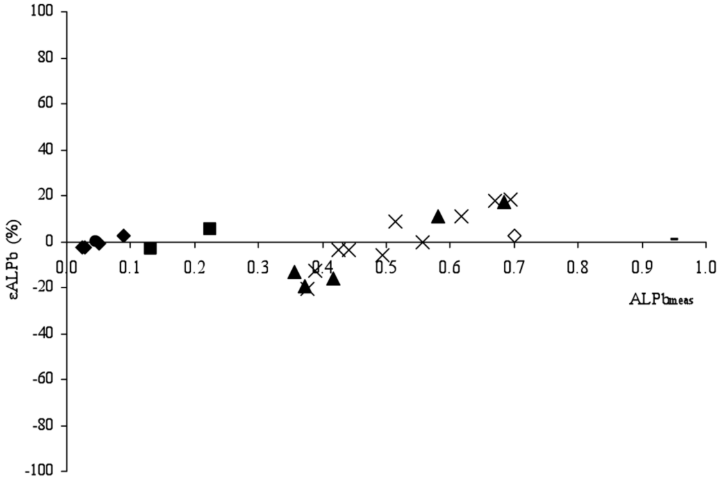


Figure 5. $ALPb_{meas}$ values vs. εALPb ($ALPb_{meas}$-$ALPb_{calc}$) expressed as %.

which takes into account the values of $ALPb_{meas}$ for the pure phases previously reported:

$$ALPb_{calc}=X_{am}{}^{\cdot 0.18}+X_{san}{}^{\cdot 0.05}+X_{cha}{}^{\cdot 0.70}+X_{phi}{}^{\cdot 0.95} \quad (1)$$

where $X_{am}$, $X_{san}$, $X_{cha}$, $X_{phi}$ represent the weight fractions of the amorphous matter, sanidine, chabazite and phillipsite, respectively.

Figure 5 reports $ALPb_{meas}$ values versus εALPb (εALPb = $ALPb_{meas}$-$ALPb_{calc}$) expressed as %. εALPb (Table 4), positive or negative, defines the deviation between $ALPb_{meas}$ and $ALPb_{calc}$. Notwithstanding this parameter is affected by the sum of the errors of the analytical methods used (RIR, Rietveld and ICP-OES) and by the increasing complexity of the investigated system characterized by the concomitant occurrence of the above considered phases, it is remarkable to evidence that all the points scatter in a well defined range equal to ±20%. On these bases it is possible to affirm that, as far as the Campanian zeolite-rich deposits are concerned, the leached Pb is substantially linked to two factors: (1) absolute concentration of the element in each single component and (2) relative concentration of the component in the rock. The highest Pb leached values are recorded for phillipsite-rich tuffs ($ALPb_{av}$=0.52). In fact, this phase is less resistant to an acid attack, if compared to chabazite-bearing samples at a parity of total Pb content, thus undergoing to a total breakdown of the framework as a consequence of the operative condition of the leaching test (de' Gennaro et al., 1983; de' Gennaro et al., 1984). This is confirmed, as already previously stated, by the behavior of the two pure zeolites for which the $ALPb_{meas}$ values are close to 1 for phillipsite and to 0.70 for chabazite.

## Conclusive remarks

Data so far discussed support the hypothesis that the leaching test described in the normative (UNI, 2008), although substantially valid in terms of procedure correctness thus assuring cautionary lead values, is definitely not suitable for zeolite-rich tuffs as temperature (~ 100 °C) and pH (0< pH <1) of the environment define the partial or, in some instances, the total framework breakdown of the zeolitic component, namely the rock constituents of major zootechnical interest. Actually, this condition likely does not occur in the digestive apparatus of those animal species (swine) grown with feedstuffs added with these minerals as anticoagulants, antibinders, etc. In fact, the gastric juice of a monogastric animals, such as swines, is characterized by high contents of HCl and the acidity ranges as a function of the digestive stage: pH = 3 at empty stomach; pH = 1 during the digestion. The strong acidity is required to transform the pepsinogen in pepsin and to enable the protein digestion. The temperature roughly corresponds to that of the normal body temperature that in swine ranges between 38.5 °C and 39.5-40 °C depending on the room temperature (Becker et al., 2003 and references therein). Whether this phenomenon would occur during the digestive function of the animal it could not be possible to exploit the beneficial effects of zeolites reported in a rich literature. It is well-known in fact, the abatement of the $NH_4^+$ ion by zeolites and what this implies in terms of breeding wellness and liveableness. Further interdisciplinary studies would shed new light on the effective role of the zeolite in this field.

Moreover, it is well-known that, for a similar additive such as the clinoptilolite of volcanic origin authorized by the European Union Commission Regulation No 2148-2004, a maximum total Pb of 80 mg/kg is allowed. Also, it is ratified that, as far as the leached Pb is concerned, the limit has been infringed from 30 to 60 mg/kg. It is therefore possible to prudentially hypothesize for these materials an $ALPb_{max}$ equal to 0.75. Considering that the campanian zeolite-rich tuffs (CI, NYT and other materials with similar geological and mineralogical features) from a normative point of view are assimilated to binders, anti-caking agents and coaugulants (E 566 natrolite-phonolite) and that for this additive a Pb leaching limit equal to 30 mg/kg has been fixed, using an $ALPb_{max}$ equal to 0.75 a maximum value of total Pb close to 50 mg/kg could be allowed. On this basis and taking into account the large case records of the present investigation, from a normative point of view the Campanian zeolite-rich tuffs could be more correctly compared to the clinoptilolite of volcanic origin (IZCLINO) as both total and leached Pb values are similar to a large extent.

By contrast, this consideration would inhibit the use of other Italian zeolite-rich tuffs such as those from Latium and *Umbria* region (Central Italy) as they show total Pb values quite high and ranging between 80 and 100 mg/kg. The low ALPb values measured after the leaching tests (see Table 4) could be explained by the quite different mineralogy of these rocks that are characterized by higher contents of amorphous + sanidine along with chabazite as dominant zeolite. Amorphous and sanidine likely determine the high total lead content; contextually they are strongly resistant to the leaching test. Moreover, among zeolite phases chabazite is quite resistant as well. The sum of these effects could explain the low values of leached Pb measured for these materials.

Further deepening of these specific aspects could be carried out *in vitro* and *in vivo* tests to better understand the behavior of these materials as additive in animal feeding and their “real” interaction with gastric juice.

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