THE DESFERRIOXAMINE TEST PREDICTS BONE ALUMINIUM BURDEN INDUCED BY Al(OH)₃ IN URAEMIC PATIENTS BUT NOT MILD HISTOLOGICAL OSTEOMALACIA

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Summary

Desferrioxamine (DFO), a chelating agent of aluminium was administered to 27 uremic patients on chronic haemodialysis or haemofiltration with a minimal parenteral exposure to aluminium but taking various amounts of Al(OH)₃ for about two years. All these patients had a double bone biopsy for measurement of their aluminium content and histomorphometric evaluation.

Bone aluminium of our patients were 10 times greater than in our uremic controls. Plasma aluminium increase (Δ Al) induced by DFO correlated better than basal plasma aluminium with bone aluminium and cumulative dose of Al(OH)₃ correlated with bone aluminium and Δ Al DFO. None of the patients had florid osteomalacia and only two had traces of aluminium staining. However 16 had mild mineralisation defect as demonstrated by low mineral appositional rate. The aluminium parameters were not different between the two groups of patients with or without mild mineralisation defect.

It is concluded that the DFO test predicts bone aluminium but not mild histological osteomalacia in uremic patients moderately aluminium over-loaded with phosphate binders.

Introduction

Although Winney et al [1] have shown that basal plasma aluminium was a good predictor of clinical bone toxicity (when >200µg/L) and even in that regard a better predictor than total bone aluminium. Milliner et al [2] proposed the desferrioxamine test (DFO test) as a non-invasive test for diagnosis of histological aluminium osteomalacia. In their experience basal aluminium was <100µg/L in six of 28 cases of aluminium bone disease whereas the plasma aluminium increase induced by desferrioxamine (ΔAl DFO) was >180µg/L in 28 of 28 patients with aluminium osteomalacia and <180µg/L in three patients without this disease.

Berland et al [3] have confirmed the value of the DFO test to distinguish between patients with and without aluminium stainable deposits, regardless
of the histological presence of osteomalacia or hyperparathyroidism [2]. Simon et al [4] have suggested that the DFO test is a better predictor than basal plasma aluminium for the evaluation of total body overload with aluminium since in patients exposed mainly to oral aluminium overload, they found a correlation between the cumulative dose of Al(OH)₃ and ΔAl DFO but not with basal plasma aluminium.

Malluche et al [5] on the other hand have concluded that the DFO test has no value for the diagnosis of aluminium bone accumulation since between 12 patients having three criteria of aluminium deposition (positive Aluminon staining, increased bone aluminium content measured by atomic absorption and abnormal peak of aluminium by X-ray analysis) and 10 patients having not all these criteria, they did not find significant difference in ΔAl DFO.

Because of these opinions on the diagnostic value of the desferrioxamine test, we performed the following study in 27 uraemic patients exposed essentially to oral aluminium overload to assess the value of the DFO test in the measurement of bone aluminium content and in the diagnosis of aluminium osteomalacia as well as to assess its link with the cumulative dose of Al(OH)₃.

Patients and methods

Patients

We studied 15 patients maintained on chronic haemodialysis for 25 ± 3 months with a dialysate aluminium always <0.3µmol/L due to reverse osmosis water treatment. They were eight men and seven women of a mean age of 54 ± 4 years. Their cumulative dose of Al(OH)₃ was 1,600 ± 345g.

Twelve patients on haemofiltration for 24±4 months were also studied. The aluminium concentration of their substitution fluid always ranged 0.15—0.6µmol/L. They were six men and six women aged 67±3 years. Their cumulative dose of Al(OH)₃ was 2,740±1,000g. No difference between haemodialysis and haemofiltration was noted for any of these clinical data.

Methods

All patients accepted having two iliac bone biopsies after double labelling by tetracycline.

One biopsy was used for histomorphometry evaluation [6] and staining of aluminium by Aluminon [7]. The other biopsy was divided into two pieces after muscle particle removal for independent measurement of aluminium content, after being precisely weighed: one determination performed in Amiens by inductively coupled plasma emission spectrometry (ICPES) [8]. The other performed in Antwerp by electrothermal atomic absorption spectrometry with a graphite furnace (EAAS) [9].

The normal mean ± SD in seven non-uraemic cadavers is 0.07 ± 0.04µmol/g of fresh bone or 1.8 ± 1.1 ppm with ICPES.

The desferrioxamine test was performed by injection of 2g of DFO during the last hour of a dialysis and measurement of plasma aluminium by ICPES before
this dialysis and 44 hours later before the subsequent dialysis. Normal non uraemic plasma aluminium is less than 0.3μmol/L.

Results

Comparison of the measurements of bone aluminium by ICPES and EAAS

The correlation between the results of the two methods obtained in 22 cases is good since the correlation coefficient is 0.91, the slope of the regression line 1.075 and the origin ordinate 2.9. This relation shows that the measurement by ICPES usually overestimates the aluminium concentration in the bone. This can be explained by the fact that bone calcium content is high and that there is a known overlap of the emission rays of aluminium and calcium, a phenomenon which does not occur with atomic absorption.

Bone aluminium content

Mean bone aluminium (±SEM) is 16.5 ± 4ppm by ICPES and 12 ± 14 by EAAS. These figures are considerably higher than in bone from non uraemic cadavers (1.8 ± 1.1ppm measured by ICPES).

Nevertheless the aluminium staining of undecalcified sections showed only traces of aluminium in two patients.

Correlation of the △Al DFO with bone aluminium content

The correlation coefficient r for the regression of bone aluminium versus △Al DFO was 0.78 (p<0.001) when bone aluminium was measured by ICPES and 0.83 when it was measured by EAAS (Figures 1 and 2). These correlations are better than those with basal plasma aluminium, the r coefficient being respectively 0.49 (p<0.05) and 0.51 (p<0.05). We suggest therefore that bone aluminium (in ppm) can be evaluated indirectly by the equation obtained with EAAS data:

\[ 3.35 \times \text{△Al DFO} \text{μmol/L} \pm 3.4 \]

Correlation of bone aluminium and △Al DFO with cumulative dose of Al(OH)₃

Significant correlations were found between the cumulative dose of Al(OH)₃ and bone aluminium content (r = 0.81 for EAAS data, p<0.001 and r = 0.64 for ICPES data, p<0.01) or △Al DFO (r = 0.83 for EAAS data and r = 0.81, p<0.001 for ICPES data).

Histomorphometrical evaluation according to osteomalacia criteria

None of our patients had florid osteomalacia with increased osteoid thickness coexistent with decreased mineral appositional rate or decreased mineralisation front. All had the osteoid thickness index of Meunier in the normal range 14–22 [6]. However 16 patients had either decreased appositional rate (<0.48μg/day) and/or decreased mineralisation front (<54% of the osteoid surface) suggesting
Figure 1

Bone Al = 3.35 Al DFO 3.36
r = 0.83 n = 22 p<0.001

Figure 2

Bone Al ICPES = 1.07 Bone Al EAAS + 2.9
r = 0.91 n = 22 p < 0.0001
a mild mineralisation defect considered by Evans as osteomalacia of type II. In the 11 other patients no such mineralisation defect was present.

Comparison of the aluminium parameters between these two groups did not show any significant difference (Table I).

**TABLE I.** Comparison of the aluminium parameters in patients with and without mineralisation defect

<table>
<thead>
<tr>
<th>Aluminium parameters mean ± SEM</th>
<th>Mineralisation Defect present n = 16</th>
<th>absent n = 11</th>
</tr>
</thead>
<tbody>
<tr>
<td>Basal plasma aluminium μmol/L</td>
<td>1.4 ± 0.4</td>
<td>2.2 ± 0.5</td>
</tr>
<tr>
<td>△Al DFO μmol/L</td>
<td>2 ± 0.3</td>
<td>2.7 ± 0.5</td>
</tr>
<tr>
<td>Bone aluminium ppm</td>
<td>14.8 ± 2.5</td>
<td>17.6 ± 5</td>
</tr>
<tr>
<td>ICPES method</td>
<td>10.9 ± 3</td>
<td>13.4 ± 4</td>
</tr>
<tr>
<td>EAAS method</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

There is no significant difference between the two groups of patients:
ICPES = Inductively Coupled Plasma Emission Spectrometry
EAAS = Electrothermal Atomic Absorption Spectrometry

**Discussion**

The bone aluminium content of our patients being much higher (10 times) than that of non uraemic controls, our patients are aluminium over-loaded although much less so than the patients of Malluche or Winney (82 and 79ppm of aluminium from dry bone). Since the cumulative doses of Al(OH)₃ correlated with the bone aluminium content and the △Al DFO the over-load of our patients appears to be mainly due to the intestinal absorption of aluminium from their phosphate binders. In fact, these patients were always exposed to low aluminium concentrations in the dialysate and the substitution fluid.

Histomorphometrical studies of their bone showed that none of our patients had florid osteomalacia since none had increased osteoid thickness and only two had traces of histological stainings for aluminium. Since in none was △Al DFO greater than 180μg/L (the highest was 162μg/L) our data agree with those of Milliner [2]. However this test does not discriminate between patients with or without mild alteration of the mineralisation process, the presence of which being demonstrated by decreased mineral appositional rate and/or decreased mineralisation front without increased osteoid thickness. Bone aluminium content did not discriminate between these two groups of patients. This lack of difference of the aluminium parameters between the two groups with and without mineralisation defect, does not mean however that their aluminium overload does not have a deleterious effect on their bone. As shown in another paper of this EDTA issue [10] when the influence of the other potentially pathogenetic factors such as plasma concentrations of PTH, 1.25(OH)₂D₃, 24.25(OH)₂D₃, Mg, Ca and PO₄ are excluded by the means of a multidimensional analysis, aluminium
parameters (bone aluminium, basal plasma and to a lesser degree ΔAl DFO are negatively correlated to osteoblastic surfaces, mineral appositional rate and bone formation rate, suggesting that their aluminium over-load decreases their bone formation.

Conclusion

In uraemic patients with mild aluminium over-load induced mainly by Al(OH)₃, the DFO test can predict bone aluminium content but not the presence of mild mineralisation defect.

References

1. Winney RJ, Cowie JF, Robson JS. Kidney Int 1984; Supplement. Symposium of New Port Beach on Aluminium Related Diseases

Open Discussion

KERR (Chairman) Can you tell us how marked is the reduction in ossification in your patients compared with what you find in pre-dialysis patients?

TOLANI I am afraid I do not have the data to hand but in pre-dialysis patients there is a low appositional rate. In our multidimensional analysis the appositional rate was mainly dependent upon vitamin D metabolites. We could detect a link, let us not say an effect, between vitamin D metabolites and apposition.