

# Metal ion release from titanium alloy orthodontic mini-implants – an in vivo study

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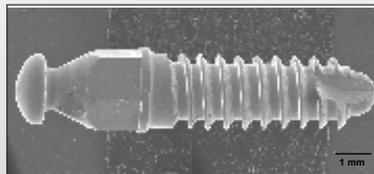
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When an implant is used to replace a missing tooth, the material of choice is the commercially pure titanium due to its proven biocompatibility, good corrosion resistance, lack of allergenicity, high specific strength, and low elastic modulus. Mini-implants are efficient as anchorage to orthodontic and orthopedic loads. In orthodontics, there is no necessity of very good integration between the implant and the host bone because the implant have to be removed at the end of the treatment. In addition, very small devices are needed for placement between the roots of the teeth, but they have to support high orthodontic loads. In that way, the material chosen to orthodontic implants is the titanium alloy (Ti-6Al-4V) due to its high strength and fatigue resistance. From hip prostheses research we know that the titanium alloy can be corroded into the human body, releasing its elements (titanium, aluminum, vanadium) to local and remote tissues, which has been associated with clinical implant failure, osteolysis, cutaneous allergic reactions, and remote site accumulation. Hip prostheses are huge prostheses, which are submitted to wear and friction, and are used for many years. On the other side, orthodontic mini-implants are smaller and are used for a short period. In addition, orthodontic mini-implants do not have surface treatment, which is usually done to enhance osseointegration, but it also prevent metal ion release. Based on this facts, is not clear if the metal ion release occurs during the orthodontic use of titanium alloy mini-implants. The purpose of this study is to measure the concentration of titanium, aluminum, and vanadium in rabbit tissues (kidney, lungs, liver, and bone) after 1, 4, and 12 weeks of insertion of titanium alloy (Ti-6Al-4V) orthodontic mini-implants.

## MATERIAL AND METHODS

### MINI-IMPLANTS



Samples of titanium alloy orthodontic mini-implants (2-mm in diameter x 6-mm in length) were supplied by Conexão Sistemas e Próteses (São Paulo, Brazil). The mini-implants were machined by turning from Ti-6Al-4V alloy, ultrasonically cleaned in acetone for 5 min, rinsed in deionized water, dried, passivated in nitric acid per ASTM F-86 at room temperature, and sterilized in an autoclave at 121 °C for 30 min.

### ANIMALS

Twenty three adult male New Zealand rabbits (3 kg) were provided and maintained by the Oswaldo Cruz Institute (Rio de Janeiro, Brazil). The animals were caged individually, fed with appropriate ration (Nuvilab rabbits) and water ad libitum. The experimental protocol was approved by the Laboratory Animal Ethics Committee of the Oswaldo Cruz Institute. The rabbits were divided in four group, according to the time they stayed with the mini-implants.

Group	Animals	Mini-implants	Period
1-week	6 rabbits	4 x 6 = 24	1 week
4-weeks	6 rabbits	4 x 6 = 24	4 weeks
12-weeks	6 rabbits	4 x 6 = 24	12 weeks
Control	5 rabbits	0	0

### ANESTHESIA

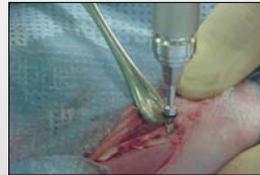


Anesthesia was induced by an intramuscular injection of tiletamine chloride (5 mg/kg) and zolazepam chloride (5 mg/kg). Hair on the anterior surface of the left tibia was clipped with electric animal clippers. Skin was cleansed with 70 % alcohol solution. A facial mask was placed to constant inhalation of oxygen and halothane to maintain general anesthesia.

### SURGICAL TECHNIQUE



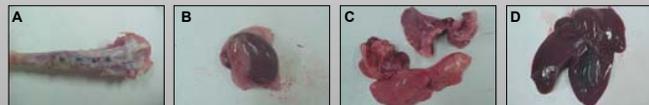
The soft tissues were incised in the long axis of the tibia. Four holes were prepared in the bone with surgical spherical burs (1.5 mm in diameter) using low rotary drill speeds (not exceeding 2,000 rpm) followed by profuse cooling with saline solution.



The implants were placed at a distance of 5 mm apart, screwed with an insertion key, and the soft tissues were sutured.



### SAMPLE OBTAINMENT

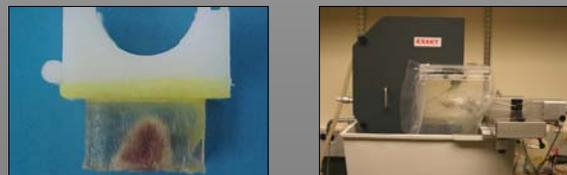


After the established times the rabbits were euthanized by exsanguination. The left tibia (A), kidneys (B), lungs (C) and liver (D) were taken. The samples were prepared according the method of analysis.

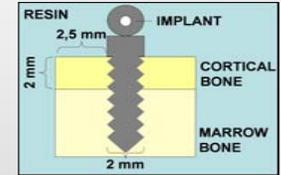
### SRXRF SPECTROMETRY



The region of the tibia containing the implants was cut in four pieces (1 mini-implant and about 2,5 mm of bone in each side). Each piece was immersed in paraformaldehyde, dehydrated in alcohol solutions and embedded in self-curing resin (Technovit 7100).

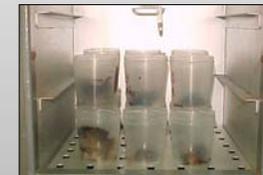


Longitudinal sections were cut with a Exakt sawing machine, ground to a final thickness of about 50 µm and mounted on carbon plates. Synchrotron radiation X-ray fluorescence spectroscopy was used for elemental analysis and mapping of the hard tissue adjacent to the mini-implants.



### ATOMIC ABSORPTION SPECTROMETRY

The selected organs (kidneys, liver, and lungs) were weighed, washed with deionized water, and stored in a freezer at -30 °C for 24 hours.



The samples were placed in a stove at 60 °C for 48 hours to initial dehydration, punched to make them homogenous, and kept in the stove at 60 °C for 7 days, until total dehydration.



Three samples of 0.5 g of each tissue were weighed, and placed in a stove at 400 °C for 5 days for sample calcination.



Three samples of 0.5 g of each tissue were weighed, and placed in a stove at 400 °C for 5 days for sample calcination. The resulting powders were digested with 65 % nitric acid (HNO<sub>3</sub>). The titanium, aluminum and vanadium content were measured by graphite furnace atomic absorption spectrometry (AAS) with background correction by transverse microprocessor modulated bipolar Zeeman magnetic field.

### STATISTICAL ANALYSIS

Statistical analysis will be performed to obtain mean and standard deviation for each tissue and metal. For significance of weight difference the data will be analyzed by 1-way ANOVA test followed by post hoc test of Tukey (confidence interval of 5%).