Introduction
The peri-implant pH value is assumed to change after the implantation of biodegradable magnesium implants or the dissolution of its corrosion products [1]. However, the peri-implant pH needs to be controlled to obtain an appropriate foreign body reaction, since the local pH value can influence the healing phase [2]. Mg(OH)\textsubscript{2} seems to have high potential for scaffolds in regenerative bone reconstruction [3], while its influence on the local tissue pH during its dissolution is not clear. Common approaches to measure the pH \textit{in vivo} are using wired techniques [4], which are limited by the use of wires. Therefore, this study investigates the pH changes around Mg(OH)\textsubscript{2} \textit{in vivo} using a pH sensitive fluorescent dye which emission spectra is changing according to the environmental pH. The emission spectra are recorded using an \textit{in vivo} imaging system which is capable to analyze the spectral information.

Materials and Methods
The pH dependent shift in the emission spectra of 5-(and-6)-carboxy SNARF\textsuperscript{®}-1 (Ex: 488-530 nm) results in two peaks which can be correlated to an acidic (600 nm) or a basic milieu (650 nm). Therefore, ratiometric measurements (Em: 600 / 650 nm) can be correlated to a pH value in a certain range. This range was investigated \textit{in vitro} and \textit{in vivo}. For \textit{in vitro} evaluation, well plates with different concentrations of SNARF\textsuperscript{®}-1 and standard buffers (pH3-10) were measured using the Maestro\textsuperscript{TM} System, CRi. For \textit{in vivo} evaluation, adult female hairless but immunocompetent mice (Crl:SKH1-hr) were used and SNARF\textsuperscript{®}-1 was administrated intravenously. The pH standardization was used to determine the pH value around a subcutaneous Mg(OH)\textsubscript{2} implant.

Results
The ratiometric measurements of 5-(and-6)-carboxy SNARF\textsuperscript{®}-1 can distinguish between acidic (pH 3-6), neutral (pH 7) and alkaline environments (pH 8-10) \textit{in vitro} as well as \textit{in vivo} (Table 1). The environment around a compressed Mg(OH)\textsubscript{2} tablet was determined as alkaline (>pH 8, Fig.1) for the initial implantation period, while the pH seem to return to neutral values after a few days and only peak into the alkaline range when the Mg(OH)\textsubscript{2} partly dissolves.

Table 1: Ratiometric values (n>3) obtained from emission spectra measured from pH standards \textit{in vitro} and \textit{in vivo}.

<table>
<thead>
<tr>
<th>pH</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
<th>10</th>
</tr>
</thead>
<tbody>
<tr>
<td>\textit{In vitro}</td>
<td>1.8</td>
<td>1.8</td>
<td>1.5</td>
<td>0.5</td>
<td>0.2</td>
<td>0.2</td>
<td>0.2</td>
<td></td>
</tr>
<tr>
<td>\textit{In vivo}</td>
<td>6.5</td>
<td>3.4</td>
<td>2.3</td>
<td>0.3</td>
<td>0.3</td>
<td>0.15</td>
<td>0.14</td>
<td>0.15</td>
</tr>
</tbody>
</table>

Fig. 1. An alkaline pH was measured \textit{in vivo}.

Discussion and Conclusions
The use of the fluorescent dye SNARF\textsuperscript{®}-1 in combination with the Maestro System\textsuperscript{TM} has provided a wireless option to measure pH values in the environment of subcutaneously implanted biomaterials in mice. The tissue pH around dissolving Mg(OH)\textsubscript{2} is turning alkaline depending on its degradation. Thus its degradation profile needs to be designed before it can be use as bone scaffolds.

References

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Disclosures
The authors have nothing to disclose.