Effects of three different embedding media on the accuracy of different electronic apex locators: An in vitro study

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Abstract
Aim: In this study, we aimed to evaluate in vitro the measurement accuracy of three electronic apex locators (EALs) in three different embedding media.

Material and Methods: Thirty maxillary central incisors were included (N = 30). The specimens were decoronated at cement–enamel junction, and the soft tissues in root canals were removed. The samples were randomly divided into three groups (n = 10). The specimens were embedded in alginate (group A), gelatin (group B) and agar agar (group C), leaving the coronal 2 mm of teeth exposed. Electronic lengths (ELs) of root canals of samples were measured using Raypex 6 (VDW, Munich, Germany), Apex ID (SybronEndo, Glendora, USA) and Ipex 2 (NSK Inc., Kanuma, Japan). In all groups, actual length (AL) values subtracted from EL values, and EL–AL values were recorded.

Results: The EL–AL values for each embedding media were classified within an error range of ± 0.5 mm and ± 1 mm, and the chi-square (χ²) test was used to compare the percentage of acceptable measurements of three EALs (α = 0.05). The measurements of Raypex 6 in agar agar medium were statistically different compared with those in alginate and gelatin media, within the error margin of ± 0.5 mm (p < 0.05). Both in alginate and gelatin media, no statistically significant difference was observed in percentages of acceptable measurements with Raypex 6, Apex ID and Ipex 2 within two ranges of error (p > 0.05).

Discussion: Within the limitations of this study, alginate and gelatin can be used safely as embedding media in studies investigating the in vitro measurement accuracy of EALs.

Keywords
Apical Foramen, Root Canal, Alginate
Introduction
The success of root canal treatment (RCT) is closely linked to the accurate determination of the working length [1]. Therefore, various ideas have been put forward to establish the apical termination of root canal preparation (RCP) and root canal filling. Theoretically, the cemento–dental junction (CDJ) has been proposed as the ideal apical limit of the RCP, which is the major exit point of the root canal system (RCS), and is also a structural and biological link between cementum and dentin in the roots of the teeth [2]. However, the CDJ is practically indistinguishable on clinical examination and can only be detected in histological sections, and the lower border of the CDJ shows wide variations within the root canal lumen [3]. Therefore, the apical constriction, which is the narrowest diameter of the root canal in the apical portion of the RCS, seems to be a more plausible anatomical landmark for the endpoint of endodontic treatment [4]. Radiographic methods have some limitations in determining the working length of RCP, such as lack of depth perception of two-dimensional vision, visual artifacts and superpositions in the images, inability to fully evaluate the variances in apical root anatomy [5], and radiation exposure. Contemporary electronic apex locators (EALs) have been introduced to overcome the limitations of radiographic methods, and most of these devices can locate the apical constriction with high accuracy [6, 7]. In previous studies, the measurement accuracy of different EALs has been extensively studied, both in vivo [8, 9] and in vitro [10, 11]. In vitro studies of EALs were performed using different electroconductive embedding media to simulate clinical conditions [12, 13]. These embedding media complete the required electrical circuit while mimicking periradicular tissues that adhere to the tooth roots and support the teeth. The effects of various embedding media, such as agar agar [14], gelatin [15] and alginate [16], on the accuracy of EALs have been investigated in previous studies. Therefore, this study aims to compare the effects of three tooth embedding media on the accuracy of three EALs.

Material and Methods
Thirty extracted human maxillary central incisors were used in the study. Care was taken to ensure that the selected teeth were single-rooted and had normal canal anatomy. Teeth with curved and calcified root canals and teeth with excessive tissue loss were excluded from the study. Apical patency was established by inserting a size 10 K-file (Dentsply Maillefer, Ballaigues, Switzerland) into the root canal until its tip was visualized in the apical foramen. The crowns of all samples were removed at the cement–enamel junction with a low-speed diamond saw (Metkon Instruments Inc., Bursa, Turkey) to set a standard reference point to determine the working length. Coronal flaring was achieved, and soft tissues in the RCS were removed using WaveOne Gold (tip 25, 0.07° taper; Dentsply Sirona, Ballaigues, Switzerland). The root canals were irrigated with 5 mL of 2.5% NaOCl (Werax, Izmir, Turkey) during the RCP. Prior to electronic measurement, the actual lengths (ALs) of root canals were determined using a size 25 K-file (Dentsply Maillefer) until its tip became visible from the apical foramen under a dental observation microscope (Carl Zeiss Gmbh., Jena, Germany) at 10× magnification. The distance between the file tip and the stopper was measured with digital calliper (Insyze Mini Digital Calliper; Istanbul, Turkey) with an accuracy of 0.01 mm. The samples were randomly divided into three groups of 10 samples each (n = 10). In group A, the specimens were embedded in alginate, with the coronal 2 mm of teeth left exposed. The electronic lengths (ELs) of the root canals of the samples were measured using Raypex 6 (VDW, Munich, Germany), Apex ID (SybronEndo, Glendora, USA) and Ipex 2 (NSK Inc., Kanuma, Japan). In groups B and C, specimens were embedded in gelatin and agar agar, respectively, and all procedures were repeated in the same manner as those in group A. In all groups, AL values were subtracted from EL values, and EL–AL values were recorded.

Statistical Analysis
All statistical analyses were performed using statistical software SPSS 22.0 (SPSS Inc., Chicago, IL, USA) (α = 0.05). The normality of the data was evaluated using the Shapiro–Wilk test, which revealed that all data were normally distributed (p > 0.05). Descriptive statistics (mean and standard deviation) of the EL–AL values for the different EALs tested in different embedding media were calculated (Table 1). The EL–AL values for each embedding media were classified within the error range of ± 0.5 mm and ± 1 mm. The chi-square (χ2) test was used to compare the percentage of acceptable measurements of the three EALs (α = 0.05).

Results
Table 1 shows the success rates of the three EALs in different embedding media within two ranges of error. The measurements of Raypex 6 in the agar agar medium were statistically different compared with those in the alginate and gelatin media, within the error margin of ± 0.5 mm (p < 0.05).

Table 2. Percentage of measurement accuracy of three apex locators in three different embedding media within two ranges of error.

<table>
<thead>
<tr>
<th>Embedding Medium</th>
<th>Apex Locator</th>
<th>Margin of Error</th>
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<tbody>
<tr>
<td></td>
<td></td>
<td>± 0.5mm (%)</td>
</tr>
<tr>
<td>Alginate</td>
<td>Raypex 6</td>
<td>90</td>
</tr>
<tr>
<td></td>
<td>Apex ID</td>
<td>90</td>
</tr>
<tr>
<td></td>
<td>Ipex 2</td>
<td>100</td>
</tr>
<tr>
<td>Gelatin</td>
<td>Raypex 6</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>Apex ID</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>Ipex 2</td>
<td>80</td>
</tr>
<tr>
<td>Agar Agar</td>
<td>Raypex 6</td>
<td>40</td>
</tr>
<tr>
<td></td>
<td>Apex ID</td>
<td>100</td>
</tr>
<tr>
<td></td>
<td>Ipex 2</td>
<td>90</td>
</tr>
</tbody>
</table>

* Superscript indicates the statistical difference compared with the values of other EALs (p < 0.05).
Embedding media effect on electronic apex locators

When using the error range of ± 0.5 mm in the agar agar test medium, the percentage of acceptable measurements of Raypex 6 was statistically significantly lower than that of Ipex 2 and Apex ID (p < 0.05). Both in the alginate and gelatin groups, no statistically significant difference was observed in the percentages of acceptable measurements of Raypex 6, Apex ID and Ipex 2 within two ranges of error (p > 0.05).

Discussion

Electroconductive media are needed to simulate periradicular tissues in in vitro studies of EALs. The most commonly used embedding media are alginate [17], gelatin [1], agar agar [18, 19] and saline solution [20]. In Baldi et al.’s study [12], saline solution was found to be the most unfavorable embedding medium that interfered with the accuracy of EALs. This is why we did not include the saline solution in the current study.

According to Gordon et al. [21], apical constriction is located at an average distance of 0.5 mm from the apical foramen. Thus, the actual working length was calculated by visualizing the tip of a size 10 K-file through the apical foramen of each sample and then subtracting 0.5 mm from the length of the file. This method has been used in previous studies [13, 22]. During RCP, 2.5% NaOCl is one of the most frequently used root canal irrigation solutions. Several studies have reported that NaOCl did not affect the determination of working length using EALs [18, 23]. Thus, we used 2.5% NaOCl as the root canal irrigant in the present study.

In a previous study [13], high accuracy was demonstrated in the measurements of alginate-embedded samples using the Raypex 5, whereas the measurement accuracy of Dentaport ZX in alginate-embedded samples was statistically significantly low. In our study, the group A samples were embedded into alginate mass, and no statistically significant difference was found in the accuracy of the three EALs within two error margins (± 0.5 mm and ± 1 mm). These favorable results may be attributed to the fact that the working principle of all three EALs relies on two-frequency basis. Using the error range of ± 0.5 mm, the accuracy rates of the EALs were 90% for Raypex 6, 100% for Ipex 2 and 90% for Apex ID. Minor differences in the success rate of these EALs may be related to the variances in the apical anatomy of the samples; these minor differences were not statistically significant.

For Root ZX in gelatin-embedded samples, Baldi et al. [12] found an accuracy rate of 60% within the ± 0.5 mm range of error and 96.7% within the ± 1 mm range of error. In our study, all group B samples were embedded in gelatin, and there was no statistical difference in the measurement accuracy of the three EALs in the gelatin group (p > 0.05). The high-accuracy measuring capability of the contemporary EALs used in this study may be related to the working characteristics of these EALs using more than one frequency signal and the calculation manner of these EALs, which rely on impedances rather than resistances.

Marroquin et al. [19] investigated the effect of the size of root canal instruments on the accuracy of EALs using agar agar-embedded teeth samples. In the aforementioned study, although no statistically significant difference was found in the success rate of the EALs for the working length determination, the most accurate results were obtained using the Raypex 5 [19]. In the current study, when using the ± 0.5 mm range of error, the lowest success rate was found using Raypex 6 (40%) among the three EALs in teeth embedded in agar agar medium (p < 0.05). The conductivity of an electrolyte is affected by the density and mobility of the charge carriers [24]. A higher quantity of mobile charge carriers in a polymer electrolyte occurs as a result of the interaction between the salt and the polymer host [25]. Based on this information, the statistically significant lower accuracy rate of agar agar as an embedding medium may be associated with the unpredictable chemical properties of the agar molecule. No statistical difference was found in the measurement accuracy between Ipex 2 and Apex ID within a ± 0.5 mm range of error in the agar agar test medium (p > 0.05).

This suggests that the technical specifications of Raypex 6 may not be suitable for use in agar agar test medium. Our study revealed that alginate and gelatin could be used safely with contemporary EALs as embedding media. However, there is a need for new studies to reassess the reliability of agar agar as a test medium, performed with modern EALs.

Scientific Responsibility Statement

The authors declare that they are responsible for the article's scientific content including study design, data collection, analysis and interpretation, writing, some of the main line, or all of the preparation and scientific review of the contents and approval of the final version of the article.

Animal and human rights statement

All procedures performed in this study were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. No animal or human studies were carried out by the authors for this article.

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Conflict of interest

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