Quantification of Tissue Shrinkage and Dehydration Caused by Microwave Ablation: Experimental Study in Kidneys for the Estimation of Effective Coagulation Volume

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ABSTRACT

Purpose: To quantify the extent of tissue shrinkage and dehydration caused by microwave (MW) ablation in kidneys for estimation of effective coagulation volume.

Materials and Methods: MW ablations were carried out in ex vivo porcine kidneys. Six study groups were defined: groups 1A, 2A, and 3A for MW ablation (90 W for 5 min, 7.5 min, or 10 min), and groups 1B, 2B, and 3B for control (without MW ablation). Pre- and postinterventional volume analyses were performed. Effective coagulation volumes (original tissue included in coagulation) were determined. Postinterventional dehydration analyses were performed with calculation of mean mass fractions of water.

Results: Mean deployed energies were 21.6 kJ ± 1.1 for group 1A, 29.9 kJ ± 1.0 for group 2A, and 42.1 kJ ± 0.5 kJ for group 3A, and were significantly different (P < .0001). Differences between pre- and postinterventional volumes were −3.8% ± 0.6 for group 1A, −5.6% ± 0.9 for group 2A, and −7.2% ± 0.4 for group 3A, and −1.1% ± 0.3 for group 1B, −1.8% ± 0.4 for group 2B, and −1.1% ± 0.4 for group 3B. Postinterventional volumes were significantly smaller than preinterventional volumes for all groups (P < .01). Underestimations of effective coagulation volume from visualized coagulation volume were 26.1% ± 3.5 for group 1A, 35.2% ± 11.2 for group 2A, and 42.1% ± 4.9 for group 3A, which were significantly different (P < .01). Mean mass fractions of water were 64.2% ± 1.4 for group 1A, 63.2% ± 1.7 for group 2A, and 62.6% ± 1.8% for group 3A, with significant differences versus corresponding control groups (P < .01).

Conclusions: For MW ablation in kidneys, underestimation of effective coagulation volume based on visualized coagulation volume is significantly greater with greater deployed energy. Therefore, local dehydration with tissue shrinkage is a potential contributor.

ABBREVIATIONS

AKS = appearance of kidney surface, MW = microwave, RF = radiofrequency

Radiofrequency (RF) and microwave (MW) ablation are emerging technologies in the field of focal tumor ablation (1,2). For the treatment of renal-cell carcinoma, thermal ablation is established as an alternative to surgical resection in selected patients (3,4). Most widely available is RF ablation, which can show oncologic results comparable to those of surgery (3). MW ablation was introduced recently to provide potential advantages compared with RF ablation: faster coagulation with more homogenous and better predictable coagulation zones (2,4,5). This can be attributed to the different energy sources between RF ablation (ie, electric current)
and MW ablation (ie, electromagnetic waves) (6). Contrary to RF ablation, dedicated clinical ablation protocols are not established for MW ablation (1,3,7).

The coagulation zone must cover tumor plus a safety margin; however, viable tissue beyond the safety margin must be protected. Right after intervention, the coagulation zone can be imaged with contrast-enhanced computed tomography (CT) to determine if the tumor plus a safety margin is completely covered. Regardless of the applied modality (eg, CT, magnetic resonance imaging, ultrasonography, or pathologic examination), the visualized coagulation zone is supposed to underestimate the extent of the original tissue included in the coagulation zone (ie, effective coagulation zone). In an experimental study, Brace et al (8) described that liver and lung tissue contracts 15%-50% during thermal ablation (8). Ganguli et al (9) described immediate renal tumor involution after RF ablation. Detailed knowledge of the extent and potential source of tissue shrinkage immediately after thermal ablation could further optimize thermal ablation procedures. If the effective coagulation zone can be quantified by using the visualized coagulation zone, a higher rate of oncologic success and a lower rate of complications may result.

Because we are aware of no such published data for the kidney, we designed the study described herein. The purpose was to quantify the extent of tissue shrinkage and dehydration caused by MW ablation in porcine kidneys. We used three different clinically relevant ablation protocols, theoretically resulting in three different amounts of deployed energy. Study goals were preinterventional and postinterventional analyses of mass, volume, and surface of the kidney; estimation of the effective coagulation volume; and postinterventional analysis of dehydration. We hypothesized that, the greater the amount of deployed energy, the greater the underestimation of effective coagulation volume from the visualized coagulation volume, with local dehydration as a potential contributor.

MATERIALS AND METHODS
A total of 24 freshly harvested porcine kidneys were used. The kidneys were cut in the transverse plane in such a manner that two symmetric halves were obtained.

Ablation Protocols and Study Groups
The commercial information from the provider was used to define the MW ablation protocols. All MW ablations were carried out with the same commercially available 2,450-MHz system (Amica; Hospital Service, Rome, Italy). Six different study groups were defined, each consisting of eight halves of kidneys. Livers in groups 1A, 2A, and 3A were treated with MW ablation performed with one 14-gauge applicator at 90 W and ablation times of 5 minutes, 7.5 minutes, or 10 minutes. The expected coagulation short axes and deployed energies were 3.5 cm and 20 kJ for group 1A, 4.3 cm and 30 kJ for group 2A, and 5.0 cm and 40 kJ for group 3A. We selected this approach because a high rate of oncologic success was reported for RF ablation in kidneys with a mean deployed energy of 27.0 kJ (for a mean tumor diameter of 18.7 mm) and a mean deployed energy of 45.7 kJ (for a mean tumor diameter of 25.2 mm) (10). In each half of kidney, one MW ablation was carried out. Livers in groups 1B, 2B, and 3B served as controls (ie, no MW ablation). The term “postinterventional” is used here to refer to the controls in analogy to groups 1A, 2A, and 3A.

Experiments
Halves of kidneys underwent preinterventional mass analysis, applying a precision balance with an accuracy of 0.5 g (PCE-LSM-6000; PCE, Meschede, Germany). Preinterventional volume analysis was performed with the water displacement technique. Halves of kidneys were submerged in a plastic container completely filled with water. The overflowing water was collected and weighed. Weight and density of water (1 g/cm³) were used to calculate the volumes of halves of kidneys. Preinterventional kidney surface analysis was carried out, with descriptions of the macroscopic appearance of kidney surface (AKS) in the area around the intended applicator tract scored on a semiquantitative four-point scale (score of 1, unremarkable tissue contour/no kidney deformation, ie, shape identical to control; 2, slight tissue retraction, ie, retraction strong enough to change the major shape of the control; 3, slight kidney deformation, ie, retraction strong enough to change the major shape of the control; and 4, severe kidney deformation, ie, very strong retraction leading to a deformation of the major shape of the control). After positioning of the applicator along the long kidney axis at a depth of approximately 4 cm, the ablation cycle was started. During MW ablation, halves of kidneys were kept in a water bath to simulate a more physiologic environment around the kidneys. After completion of the ablation cycle, the applicator was removed. Postinterventional mass, volume, and kidney surface analyses (in the area around the applicator tract) were performed in analogy to preinterventional mass, volume and kidney surface analyses for all study groups. For the controls, mass, volume, and kidney surface analyses were carried out at time points comparable with groups 1A, 2A, and 3A. Finally, halves of kidneys were cut in the sagittal plane and photographed, and the photographs were digitized.

Estimation of Effective Coagulation Volume
Estimation of effective coagulation volume was performed for groups 1A, 2A, and 3A, and defined as the volume of the original tissue included in the coagulation
zone. To estimate the effective coagulation volume, the visualized coagulation volume, the shrinkage volume, and the volume-corrected shrinkage were determined. The visualized coagulation volume was defined as the area showing a change from red to white coloration during MW ablation (6,8). Coagulation long and short axes were measured by applying commercially available software (Photoshop CS3 Extended, version 10.0; Adobe Systems, San Jose, California). The coagulation long axis was defined in the direction of the applicator tract, and the coagulation short axis was defined perpendicular to the applicator tract (11,12). The visualized coagulation volume was calculated as previously described (13):

$$\text{Visualized coagulation volume} = (\pi/6) \times \text{coagulation long axis} \times (\text{coagulation short axis})^2$$

The shrinkage volume was calculated as the difference between preinterventional and postinterventional volume (differential volume). Volume-corrected shrinkage figures were calculated as the differences between differential volumes in groups 1A, 2A, and 3A and the differential volumes in the corresponding control groups. The effective coagulation volume was calculated as the sum of the visualized coagulation volume and the volume-corrected shrinkage. The percentage of visualized coagulation volume from the effective coagulation volume was calculated assuming that the effective coagulation volume was 100%. Underestimation of the effective coagulation volume from the visualized coagulation volume was calculated assuming that the visualized coagulation volume was 100%.

**Postinterventional Dehydration Analysis**

For each half of kidney, two tissue blocks with a volume of approximately 1 cm$^3$ were removed from the center (around the applicator tract), midportion (center of the white coagulation area), and periphery (transitional zone to noncoagulated tissue) of the visualized coagulation zones for groups 1A, 2A, and 3A, and from comparable locations for the control groups. The tissue blocks underwent predehydration and postdehydration mass analysis with the use of a precision balance with an accuracy of 0.01 g (KB 1200-2N; Kern and Sohn, Balingen-Frommern, Germany). Dehydration was performed for 18 hours at 75°C with a heating cabinet. The mass fraction of water was calculated as follows for the three different locations for each study group (8):

$$\text{Mass fraction of water} = \left(\frac{\text{pre-dehydration mass} - \text{post-dehydration mass}}{\text{pre-dehydration mass}}\right) \times 100\%$$

**Statistics**

Prism software (version 4.00; GraphPad, La Jolla, California) was used for statistical analysis. Data are presented as means, standard deviations, and ranges. The Wilcoxon signed-rank test was used to define statistical differences between (i) preinterventional and postinterventional data (mass, volume, and AKS score) and (ii) groups 1A, 2A, and 3A and corresponding control groups (postinterventional dehydration analysis). The Kruskal–Wallis test was used to define statistical differences between (i) groups 1A, 2A, and 3A (deployed energy, AKS score, estimation of effective coagulation volume, and postinterventional dehydration analysis) and (ii) groups 1B, 2B, and 3B (AKS score and post-interventional dehydration analysis). Linear regression analyses were used with (i) deployed energy on the $x$ axis and visualized coagulation volume on the $y$ axis, (ii) deployed energy on the $x$ axis and effective coagulation volume on the $y$ axis, and (iii) visualized coagulation volume on the $x$ axis and effective coagulation volume on the $y$ axis. The level of statistical significance was $P = .05$.

**RESULTS**

All MW ablations were carried out as planned. Mean deployed energies were 21.6 kJ ± 1.1 (19.4–23.1 kJ) for group 1A, 29.9 kJ ± 1.0 (28.6–31.5 kJ) for group 2A, and 42.1 kJ ± 0.5 (41.3–43.1 kJ) for group 3A, which were significantly different ($P < .0001$).

**Mass Analysis**

Complete mass analysis data are listed in Table 1. Differences between preinterventional and postinterventional masses were −5.0% ± 0.2 for group 1A, −6.5% ± 0.9% for group 2A, and −8.1% ± 0.6 for group 3A, compared with −1.7% ± 0.4 for group 1B, −2.3% ± 0.9 for group 2B, and −1.8% ± 0.6 for group 3B. Postinterventional masses were significantly smaller than preinterventional masses for all study groups ($P < .01$).

**Volume Analysis**

Complete volume analysis data are listed in Table 1. Differences between preinterventional and postinterventional volumes were −3.8% ± 0.6 for group 1A, −5.6% ± 0.9 for group 2A, and −7.2% ± 0.4 for group 3A, compared with −1.1% ± 0.3 for group 1B, −1.8% ± 0.4 for group 2B, and −1.1% ± 0.4 for group 3B. Postinterventional volumes were significantly smaller than preinterventional volumes for all study groups ($P < .01$).

**Kidney Surface Analysis**

For groups 1A, 2A and 3A, postinterventional AKS scores were significantly higher compared with preinterventional AKS scores (2.3 ± 0.5 vs 1.0 ± 0.0, 2.8 ± 0.7 vs 1.0 ± 0.0, and 3.8 ± 0.5 vs 1.0 ± 0.0, respectively; $P < .01$; Fig 1). Moreover, for groups 1A, 2A, and 3A, postinterventional AKS scores were significantly different (2.3 ± 0.5 vs 2.8 ± 0.7 vs 3.8 ± 0.5; $P < .01$). These results indicate tissue retraction/kidney deformation in the area around the applicator tract after MW ablation. For groups 1B, 2B, and 3B, preinterventional and postinterventional AKS scores were identical (each 1.0 ± 0.0).
Estimation of Effective Coagulation Volume

Complete estimated effective coagulation volume data are listed in Table 2. Visualized coagulation volumes were 11.7 cm$^3 \pm 1.6$ for group 1A, 14.2 cm$^3 \pm 1.7$ for group 2A, and 17.0 cm$^3 \pm 1.8$ for group 3A, which were significantly different ($P < .0001$). Effective coagulation volumes were 14.7 cm$^3 \pm 1.6$ for group 1A, 19.0 cm$^3 \pm 1.8$ for group 2A, and 24.1 cm$^3 \pm 2.6$ for group 3A, which were significantly different ($P < .0001$). Calculations of underestimation of effective coagulation volume from visualized coagulation volume were 26.1% $\pm 3.5$ for group 1A, 35.2% $\pm 11.2$ for group 2A, and 71.0% $\pm 0.0$ for group 3A, which were significantly different ($P < .0001$).

Table 1. Mass and Volume Analysis

<table>
<thead>
<tr>
<th>Group A (MW Ablation)</th>
<th>Group B (Control)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Preinterventional</strong></td>
<td><strong>Postinterventional</strong></td>
</tr>
<tr>
<td>Mass Analysis (g)</td>
<td>Mass Analysis (g)</td>
</tr>
<tr>
<td>1</td>
<td>142.0 $\pm 6.8^*$</td>
</tr>
<tr>
<td>(136.0–152.0)</td>
<td>(129.0–144.0)</td>
</tr>
<tr>
<td>2</td>
<td>146.0 $\pm 4.8^*$</td>
</tr>
<tr>
<td>(138.0–152.0)</td>
<td>(128.0–141.0)</td>
</tr>
<tr>
<td>3</td>
<td>137.1 $\pm 6.7^*$</td>
</tr>
<tr>
<td>(127.0–144.0)</td>
<td>(116.0–133.0)</td>
</tr>
</tbody>
</table>

Values presented as means $\pm$ standard deviation. Values in parentheses are ranges.

*P < .01 (Wilcoxon signed-rank test) comparing pre-interventional and postinterventional data.

MW = microwave.
for group 2A, and 42.1% ± 4.9 for group 3A, which were significantly different ($P < .01$).

**Linear Regression Analyses**
The equation for linear regression between deployed energy on the x axis and visualized coagulation volume on the y axis was $y = 0.26x + 6.21$, with an $R^2$ of 0.66 and $P < .0001$. The equation for linear regression between deployed energy on the x axis and effective coagulation volume on the y axis was $y = 0.46x + 5.07$, with an $R^2$ of 0.81 and $P < .0001$ (Fig 3). The equation for linear regression between visualized coagulation volume on the x axis and effective coagulation volume on the y axis was $y = 1.53x – 2.56$, with an $R^2$ of 0.92 and $P < .0001$.

**Postinterventional Dehydration Analysis**
Complete postinterventional dehydration data are listed in Table 3. Mean mass fractions of water were 64.2% ± 1.4 for group 1A, 63.2% ± 1.7 for group 2A, and 62.6% ± 1.8 for group 3A, which were not significantly different. Mean mass fractions of water were 78.1% ± 2.1 for group 1B, 76.8% ± 2.4 for group 2B, and 75.4% ± 2.6 for group 3B, which were not significantly different. For groups 1A, 2A, and 3A, mean mass fractions of water were significantly lower compared with corresponding controls ($P < .01$, $P < .01$, and $P < .01$, respectively). Regarding the location within the coagulation zone, mass fractions of water for groups 1A, 2A, and 3A were smallest in the center, intermediate in the midportion, and largest in the periphery.

**DISCUSSION**
The present study indicates that, for MW ablation in kidneys, underestimation of the effective coagulation volume from the visualized coagulation volume is significantly higher with larger amounts of deployed energy. As the mean mass fractions of water are significantly lower for groups 1A, 2A, and 3A compared with the corresponding controls, local dehydration is one potential contributor to the observed tissue shrinkage. Consequently, our hypothesis has been confirmed.
Tissue shrinkage and dehydration caused by RF and MW ablation has never been quantified in kidneys to our knowledge. As immediate renal tumor involution after thermal ablation was reported to be approximately 20%, and MW ablation is a new technique for the treatment of renal-cell carcinoma, the present study has potential clinical implications (4,7,9). Quantification of the extent of tissue shrinkage after MW ablation could be important for a nephron-sparing approach with complete tumor destruction.

Brace et al (8) analyzed RF and MW ablation–induced tissue shrinkage in liver and lung. Three pairs of markers were introduced into the tissue at distances of 1.0 cm, 2.0 cm, and 3.0 cm. Compared with controls, the minimum distances of pairs of markers decreased 2.9–4.8 mm after RF ablation in liver, 3.6–9.0 mm after MW ablation in liver, 5.2–14.2 mm after RF ablation in lung, and 4.9–13.7 mm after MW ablation in lung. Accordingly, the authors (8) calculated relative tissue shrinkage of 15% after RF ablation in liver, 30% after MW ablation in liver, 55% after RF ablation in lung, and 49% after MW ablation in lung. For both techniques, the extent of tissue shrinkage was more pronounced in the periphery of the coagulation zone.

Table 3. Postinterventional Dehydration Analysis

<table>
<thead>
<tr>
<th>Location within Coagulation Zone</th>
<th>Group A (MW Ablation)</th>
<th>Group B (Control)</th>
<th>P Value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Center</td>
<td>56.0 ± 5.0 (51.0–64.0)</td>
<td>77.6 ± 2.8 (74.0–81.0)</td>
<td>&lt; .01</td>
</tr>
<tr>
<td>Midportion</td>
<td>64.8 ± 1.8 (62.0–67.0)</td>
<td>78.8 ± 3.4 (74.0–84.0)</td>
<td>&lt; .01</td>
</tr>
<tr>
<td>Periphery</td>
<td>71.8 ± 2.8 (68.0–76.0)</td>
<td>77.9 ± 4.1 (74.0–81.0)</td>
<td>&lt; .05</td>
</tr>
<tr>
<td>Mean</td>
<td>64.2 ± 1.4 (62.0–66.0)</td>
<td>78.1 ± 2.1 (76.0–81.0)</td>
<td>&lt; .01</td>
</tr>
<tr>
<td>Group 2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Center</td>
<td>54.3 ± 4.5 (50.0–61.0)</td>
<td>76.1 ± 3.6 (72.0–81.0)</td>
<td>&lt; .01</td>
</tr>
<tr>
<td>Midportion</td>
<td>63.1 ± 3.0 (59.0–66.0)</td>
<td>78.5 ± 2.5 (76.0–82.0)</td>
<td>&lt; .01</td>
</tr>
<tr>
<td>Periphery</td>
<td>72.3 ± 3.2 (68.0–77.0)</td>
<td>75.9 ± 3.3 (72.0–79.0)</td>
<td>NS</td>
</tr>
<tr>
<td>Mean</td>
<td>63.2 ± 1.7 (61.3–66.0)</td>
<td>76.8 ± 2.4 (74.3–79.7)</td>
<td>&lt; .01</td>
</tr>
<tr>
<td>Group 3</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Center</td>
<td>53.5 ± 2.7 (49.0–56.0)</td>
<td>72.2 ± 3.7 (71.0–81.0)</td>
<td>&lt; .01</td>
</tr>
<tr>
<td>Midportion</td>
<td>62.4 ± 3.8 (57.0–68.0)</td>
<td>76.3 ± 4.0 (70.0–80.0)</td>
<td>&lt; .01</td>
</tr>
<tr>
<td>Periphery</td>
<td>71.9 ± 2.9 (69.0–76.0)</td>
<td>75.0 ± 2.3 (72.0–78.0)</td>
<td>NS</td>
</tr>
<tr>
<td>Mean</td>
<td>62.6 ± 1.8 (58.7–64.3)</td>
<td>75.4 ± 2.6 (71.0–78.0)</td>
<td>&lt; .01</td>
</tr>
<tr>
<td><strong>P-value</strong></td>
<td>NS</td>
<td>NS</td>
<td></td>
</tr>
</tbody>
</table>

MW = microwave, NS = not significant.
*Wilcoxon signed rank test.
†Kruskal–Wallis test for mean.
compared with RF ablation in liver. The overall higher extent of tissue shrinkage in lung was explained by a significant contribution of air-filled space collapse.

In view of the lack of published data for the kidney, the present results can be discussed in relation to the data presented for liver, and are comparable. Quantification revealed underestimation of the effective coagulation volume from the visualized coagulation volumes of 26.1%–42.1%, depending on deployed energy. In this context, the role of the mass fraction of water could be relevant. In the study of Brace et al (8), the mass fraction of water in liver was 71% before thermal ablation. After RF and MW ablation with coagulation zones of approximately 33 mm, the mass fractions of water were 56% and 51% at distances of 5 mm to the applicator, 63% and 67% at distances of 10 mm to the applicator, and 68% and 71% at distances of 15 mm to the applicator. These data are in the range of our results if coagulation zones and locations of tissue blocks used for determination of mass fractions of water are considered. In analogy to the study of Brace et al (8), the extent of tissue shrinkage in kidneys caused by MW ablation in the present study seems to be correlated with the degree of local dehydration. However, it appears likely that different organ anatomy with smaller parenchymal thickness, lower surface/volume ratio, and stronger connective tissue fixation (pelvic system) for the kidney could explain some of the differences.

Additionally, the different energy sources have to be taken into account. During RF ablation, a current circuit is closed, and moving electrons heat up the tissue via friction (14,15). During MW ablation, electromagnetic waves induce precipitation of water dipoles, resulting in frictional heat (2). Local energy transfer into the tissue is supposed to be more effective for MW ablation (9,16,17). This observation could explain the lower mass fraction of water combined with the higher extent of tissue shrinkage for MW ablation compared with RF ablation. During a more effective energy transfer, the heated tissue could lose more water through an evaporation process. Because the gas pressure increases, the water vapor could diffuse and condensate within the kidney and/or escape the system. Water evaporation as well as water vapor diffusion, condensation, and escape could be regarded as a process of water movement (18). In previous ex vivo tests, we noticed considerable smoke emission when MW ablations were performed in room air. As smoke emission could be accompanied by vapor escape, we kept halves of kidneys in a water bath in the present study. The water was intended to simulate a more physiologic environment (ie, closed system kidney) compared with room air (ie, open system kidney), for example with reference to surrounding density (air standard, 0.0012 g/cm³; water standard, 1.0 g/cm³; and human fatty tissue standard, 0.94 g/cm³). However, the thermodynamic processes are complex, and detailed assessment of phase transformations (eg, from liquid to vapor and vice versa) would go beyond the scope of one study. In the present study, the mass fractions of water for groups 1A, 2A, and 3A were lowest in the center, intermediate in the midportion, and highest in the periphery of the visualized coagulation zone. Whether—and, if so, to what extent and how—water movement took place outside of the visualized coagulation zone remains unclear (8,18). The formation of gas bubbles within the water during MW ablation, as well as swelling of noncoagulated tissue, indicates that water movement is a complex process. For clinical MW ablation, this process could be relevant for the coagulated and noncoagulated tissue as well as for the environment (18).

A limitation of the present study is the ex vivo model. Comparison of different types of tissue (eg, fibrous and fatty) could yield additional important information. Nevertheless, we think this approach is valuable because it allows a better controllable setup compared with in vivo study designs. There exist very precise techniques for the quantification of dewatering (eg, yttrium-heavy water), but, because this technique was not available in our institution, we used the gravitational dewatering method. The coagulation zone was not completely surrounded by normal tissue, and the anatomy of halves of kidneys could have affected the coagulation shape. Theoretically, MW ablation protocols can be adjusted in such a manner that the coagulation zone is elliptical and completely surrounded with normal tissue. Finally, we did not include RF ablation. In previous tests, our monopolar RF system did not show reliability for ex vivo use.

In conclusion, for MW ablation in kidneys, underestimation of the effective coagulation volume from the visualized coagulation volume is significantly greater with greater amounts of deployed energy. Local dehydration with tissue shrinkage is a potential contributor to these findings.

REFERENCES


