The endothelium plays an important role in regulating blood flow during various metabolic perturbations. Studies have shown that nitric oxide (NO) contributes, in part, to reactive hyperemia in the forearm because NO synthase inhibitors partially attenuate the forearm blood flow (FBF) response (9,14). In particular, investigators have focused on peak forearm vascular resistance (FVR) and FBF during reactive hyperemia as a measure of peak vasodilatory capacity. The primary factors contributing to peak vasodilatory capacity are local regulators such as adenosine and prostaglandin (3,8,13). Although NO may have only a small role in determining blood flow during reactive hyperemia, NO also interacts with adrenergic mechanisms.

The eNOS gene consists of 26 exons and 25 introns located on the long arm of chromosome 7 at 7q35→36 (19). In the promoter 5′ flanking region, a single nucleotide polymorphism (SNP) with a T to C substitution occurring at nucleotide position −786 (T-786C) has been identified (12,31). Recently, in vitro studies have shown that individuals with the C allele had significantly decreased promoter activity compared with individuals without a C allele (31). This mutation suppresses eNOS transcription, which is consistent with reduced NO production. In addition, a shear stress responsive element has been identified in a region near the eNOS promoter (12). These functional characteristics of the eNOS T-786C polymorphism suggest individuals with a C allele may demonstrate differences in FVR and FBF during reactive hyperemia.

Aerobic exercise training has the capacity to improve endothelial function (9,28). Two cross-sectional studies found a greater FBF response during acetylcholine infusions in healthy exercise trained individuals than their age- and sex-matched controls (6,15). Testa et al. (28) reported an increase in peak calf reactive hyperemia after 12 wk of aerobic exercise training in chronic heart failure patients. Interestingly, in this study, approximately half of the sub-
jects demonstrated an increase in eNOS gene expression whereas there was no change in the remaining subjects. This finding of heterogeneous responses in eNOS gene expression to a standardized exercise stimulus suggests that heterogeneity at the DNA level may be responsible for the range of responses. Together, these studies suggest that the eNOS gene interacts with aerobic exercise training to affect FBF. There are no studies that have determined whether the eNOS T-786C gene polymorphism interacts with physical activity levels to affect FVR and FBF during reactive hyperemia.

The purpose of the present study was to determine whether the eNOS T-786C gene polymorphism interacts with habitual physical activity level to affect FVR and FBF at rest and at 1 (peak vasodilation), 2, and 3 min during reactive hyperemia in healthy Caucasian women. We also sought to determine whether there was a main effect of eNOS genotype on FVR and FBF because the C allele has been associated with reduced eNOS gene transcriptional and promoter activity. We hypothesized that C allele carriers (TC+CC genotypes) would have a greater FVR and a lower FBF at rest and at each minute during reactive hyperemia compared with the TT genotype group. In addition, because high levels of cardiovascular fitness are associated with better peripheral vascular function, we hypothesized that that endurance-trained subjects with the TT genotype would have the lowest FVR and the greatest FBF at min 1, 2, and 3 during reactive hyperemia compared with the sedentary subjects with the CC genotype.

METHODS

Subjects. The study was approved by the University of Maryland Human Subjects Review Board. All subjects signed an informed written consent form. The subjects were part of a larger project designed to assess the effects of genotype on exercise cardiovascular hemodynamics in young females conducted in the Department of Kinesiology, University of Maryland College Park. Healthy sedentary and endurance-trained 18- to 30-yr-old Caucasian female volunteers were recruited from the University of Maryland and the surrounding metropolitan area through print and media advertisements. A total of 63 subjects (35 endurance-trained, 28 sedentary) were recruited for the study. Initially, potential subjects who responded to the advertisements were interviewed by telephone using a screening questionnaire. Women with a history of the coronary artery disease, other cardiovascular diseases, smoking, diabetes, medications affecting cardiovascular responses, or orthopedic conditions were excluded from participating in this study. In addition to their health history, the subject’s previous physical activity level was recorded to preliminarily group them as sedentary or endurance-trained. Sedentary requirements were regularly participating in any aerobic exercise <20 min·d⁻¹ less than 3 d·wk⁻¹ and endurance-trained requirements were participating in aerobic exercise 45–60 min·d⁻¹ at least 5 d·wk⁻¹ at a moderate to high intensity for a minimum of 2 yr.

During the first screening visit, subjects completed a detailed medical history questionnaire to verify the telephone interview information. A 15-mL blood sample was then obtained by venipuncture from the antecubital vein for DNA analysis. Finally, the subject’s height, weight, and skinfold measurements were recorded. The Jackson and Pollock (1) seven-site skinfold technique was used to determine body composition.

Maximal oxygen consumption. The subject’s maximal oxygen consumption (\(\dot{V}O_2\) max), a surrogate measure of habitual physical activity levels, was assessed utilizing a maximal treadmill exercise test as previously described (5). Throughout and at the termination of the test, blood pressure and heart rate were monitored and recorded. A true maximal test was based on standard criteria if two of the following criteria were met: RER >1.10, heart rate ≥ (220 – age), and a plateau in \(\dot{V}O_2\) during the last stage of the test (<150 mL·min⁻¹·kg⁻¹ increase in \(\dot{V}O_2\)) (5). If the criteria were not met, the subject returned on a separate day to repeat the test.

Forearm blood flow. All studies were conducted between 7:00 and 9:00 a.m. after an overnight fast of 12 h. All subjects were studied between the 2nd and 7th day of their menstrual cycle when estrogen levels were at the lowest and have the least affect on endothelial function. Subjects assumed the supine position for 15 min. After the 15-min rest period, FBF was measured using venous occlusion plethysmography and a mercury-filled silastic strain gauge placed around the widest part of the subject’s nondominant forearm. The FBF response was recorded on a strip chart and calculated using the slope of the strain gauge output. Twenty seconds before each FBF measurement, the wrist cuff was inflated to 200 mm Hg to remove hand blood flow from the FBF measurement. Three successive intermittent FBF measurements were taken at baseline by inflating the arm cuff to 55 mm Hg using an automatic rapid cuff inflator. FBF was recorded for seven heartbeats. In addition, resting heart rate and blood pressure were recorded simultaneously in the contralateral arm.

Reactive hyperemia, the vasodilatory response to transient vascular occlusion, was used to increase FBF. To induce ischemia and subsequent reactive hyperemia, the upper arm cuff was inflated to 50 mm Hg above the subject’s resting systolic blood pressure obtained during the 15-min rest period at baseline for 5 min (16). After the upper arm cuff was deflated, FBF was measured intermittently each min for 3 min. Studies have shown that tissues appear to recover from hypoxia within 3 min and that peak blood flow occurs at approximately 1 min (4). FBF was calculated as FBF (mL·100 mL⁻¹·FAV⁻¹·min⁻¹) = K × dV/dT where \(K\) is a constant, \(V\) is voltage, \(T\) is time, and FAV is forearm volume. FVR was calculated as the mean arterial pressure (MAP) divided by the corresponding FBF and reported as arbitrary units (u).

eNOS Genotyping. Genomic DNA was extracted from the leukocytes of the blood sample utilizing a TrueGene kit (Gentra Systems, Minneapolis, MN). Subjects were genotyped for the eNOS −786 site using polymerase chain reaction amplification with flanking primers F:5'-CAC-CCAGGCCCCACCCCAACT-3' and R:5'-GCCGCAGGTC-GACAGAGAGACT-3'. DNA was denatured for 5 min at
TABLE 1. Subject characteristics in the genotype × physical activity level groups.

<table>
<thead>
<tr>
<th>Variables</th>
<th>Sed TC+CC (N = 13)</th>
<th>End Tr TC+CC (N = 18)</th>
<th>Sed TT (N = 15)</th>
<th>End Tr TT (N = 17)</th>
<th>ANOVA P Values</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (yr)</td>
<td>24.4 ± 1</td>
<td>25.0 ± 1</td>
<td>23.6 ± 1</td>
<td>26.8 ± 1</td>
<td>0.26</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>161.9 ± 2</td>
<td>162.4 ± 1</td>
<td>165.4 ± 2</td>
<td>165.0 ± 2</td>
<td>0.91</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>60.1 ± 4</td>
<td>60.0 ± 1</td>
<td>66.5 ± 4</td>
<td>59.3 ± 2</td>
<td>0.21</td>
</tr>
<tr>
<td>Body fat (%)</td>
<td>25.5 ± 2</td>
<td>20.0 ± 1</td>
<td>26.5 ± 2</td>
<td>18.5 ± 1</td>
<td>0.33</td>
</tr>
<tr>
<td>Fat free mass (kg)</td>
<td>44.1 ± 1</td>
<td>47.8 ± 1</td>
<td>48.5 ± 2</td>
<td>48.3 ± 1</td>
<td>0.20</td>
</tr>
<tr>
<td>VO₂max (mL kg⁻¹ min⁻¹)</td>
<td>34.1 ± 1</td>
<td>44.5 ± 1</td>
<td>33.3 ± 1</td>
<td>46.2 ± 1</td>
<td>0.34</td>
</tr>
<tr>
<td>Baseline HR (beats min⁻¹)</td>
<td>66.0 ± 3</td>
<td>57.7 ± 2</td>
<td>64.6 ± 3</td>
<td>56.8 ± 2</td>
<td>0.91</td>
</tr>
<tr>
<td>Baseline SBP (mm Hg)</td>
<td>110.9 ± 3</td>
<td>110.4 ± 2</td>
<td>109.8 ± 3</td>
<td>115.5 ± 5</td>
<td>0.45</td>
</tr>
<tr>
<td>Baseline DBP (mm Hg)</td>
<td>72.6 ± 2</td>
<td>68.6 ± 2</td>
<td>71.5 ± 2</td>
<td>72.9 ± 2</td>
<td>0.23</td>
</tr>
<tr>
<td>Baseline MAP (mm Hg)</td>
<td>85.3 ± 2</td>
<td>82.7 ± 2</td>
<td>94.2 ± 2</td>
<td>87.0 ± 3</td>
<td>0.28</td>
</tr>
</tbody>
</table>

VO₂max: maximal oxygen consumption; HR, heart rate; SBP, systolic blood pressure; DBP, diastolic blood pressure; MAP, mean arterial pressures; Sed, sedentary; End Tr, endurance trained. Values are means ± SE.

95°C followed by 35 cycles of denaturation (30 s, 95°C), annealing (15 s, 54°C), and extension (30 s, 72°C). The amplicon was digested overnight at 37°C using 5 μM of MspI followed by electrophoresis for 4 h in a gel composed of 2% agarose + 1% Nusieve (FMC, Inc.). The T allele yields a fragment of 415 bp, and the C allele yields fragments of 370 bp and 45 bp.

Data and statistical analysis. In order to detect a 15% difference in FBF at a power of 0.80 and an alpha level of 0.05, we calculated that a minimum of 15 subjects were required in each genotype group. Therefore, based on the reported C allele frequency, the C allele carriers were grouped into a combined TC+CC genotype group and compared with the TT genotype group (25,33). Descriptive statistics were calculated for the four genotype × physical activity level groups (sedentary TC+CC, endurance-trained TC+CC, sedentary TT, and endurance-trained TT) using two-way ANOVA with eNOS genotype and physical activity level as the independent variables. Because body composition and resting FBF can influence the hemodynamic response during reactive hyperemia, all subsequent analyses were covaried for percent body fat and baseline FBF. Time-course changes in FBF during reactive hyperemia were compared using a three-way ANOVA (genotype × physical activity level × time) with repeated measures. A two-way ANOVA with eNOS genotype and physical activity level used as independent variables was used to assess potential interactive and main effects between the dependent variables at rest (baseline) and at 1, 2, and 3 min during reactive hyperemia. The primary dependent variables were FVR and FBF at baseline and at min 1 (peak vasodilation), 2, and 3 during reactive hyperemia. Additional dependent variables were the percent changes in FVR and FBF at min 1, 2, and 3. To compare differences between the four genotype × physical activity level groups, post hoc comparisons were performed using the Bonferroni procedure. The data was analyzed using StatView 5.0 (SAS Institute Inc.). All results are presented as means ± SE. A value of P < 0.05 was considered statistically significant.

RESULTS

Subject characteristics. The subject characteristics of the eNOS T-786C genotype groups and the physical activity level groups are summarized in Table 1. There were 63 women in two eNOS genotype groups (TT genotype, N = 32; TC+CC genotype, N = 31) and 63 women in two physical activity level groups (sedentary, N = 28; and endurance trained, N = 35). This resulted in the following eNOS genotype × physical activity levels groups (sedentary TC+CC, N = 13; endurance-trained TC+CC, N = 18; sedentary TT, N = 15; and endurance-trained TT, N = 17). There were no differences in subject characteristics between the four genotype × physical activity levels groups. As expected, VO₂max was significantly greater (P < 0.0001), and percent body fat (P < 0.001) and resting heart rate (P = 0.002) were significantly lower in the endurance-trained women than in the sedentary women. There were no significant differences in age, height, weight, and blood pressure between the two physical activity level groups.

In general, the sedentary TC+CC group had the greatest baseline FVR and the lowest baseline FBF compared with the other three groups (Table 2). Within the entire group of subjects and within each of the four genotype × physical activity level groups, FBF increased significantly (P < 0.0001) during reactive hyperemia compared with the baseline values. After accounting for differences in baseline FBF and percent body fat, the greatest FBF at 1 min (peak vasodilation) was demonstrated by the sedentary TT group and the lowest peak FBF was observed in the sedentary TC+CC group. In all groups, FBF remained significantly higher than the baseline values at min 2. At min 3, FBF in all groups except the endurance-trained TT group remained higher than the baseline values. Systolic blood pressure in all groups did not change; however, diastolic blood pressure tended (P = 0.09) to decrease in all groups during the reactive hyperemic period.

Interactive effect of the eNOS T-786C gene polymorphism and physical activity level on hemodynamic variables. Table 2 summarizes the interactive effects between eNOS T-786C genotype and physical activity level on hemodynamic variable at baseline and during reactive hyperemia. All analyses used baseline FBF and percent body fat as covariates, except when analyzing baseline FBF. In this case, only percent body fat was used as a covariate. There was a significant interactive effect between eNOS genotype and physical activity level on baseline FVR (P = 0.0003) and baseline FBF (P = 0.03). This interaction resulted because within the sedentary group, the TT genotype had the lowest baseline FVR, but within the endurance-
trained group, the TC+CC and the TT genotype groups had identical baseline FVR values. The differences in FVR were not related to differences in MAP. Within the sedentary group, the TT genotype had greater baseline FBF than the TC+CC genotype group; however, within the endurance-trained group, the TC+CC genotype group had a greater baseline FBF than the TT genotype group. There was also a significant interactive effect between eNOS genotype and physical activity level on FVR at min 2 ($P = 0.04$) and a tendency for an interactive effect at min 3 ($P = 0.07$) during reactive hyperemia. Within the sedentary group, the TT genotype group had the lowest FVR, but within the endurance-trained group, the TC+CC genotype group had the lowest FVR at 2 min. There were no interactive effects with respect to any of the blood pressure variables.

**Main effect of the eNOS T-786C gene polymorphism and physical activity level on hemodynamic variables.** Table 2 summarizes the main effects of physical activity level and eNOS genotype. There was only one main effect of physical activity level detected. The sedentary subjects had a greater baseline FVR compared with the endurance-trained subjects ($41 \pm 3$ vs $35 \pm 2$, $P = 0.0003$). All of the other main effects identified were related to the eNOS T-786C genotype, and they were all observed in variables related to peak vasodilation measured at the first minute after arterial occlusion. The TT genotype group had a greater peak FBF compared with the TC+CC genotype group ($7.0 \pm 0.3$ vs $5.9 \pm 0.4$ mL·100 mL$^{-1}$·FAV·min$^{-1}$, $P = 0.03$). This corresponded to the tendency for the TT genotype group to have lower FVR at 1 min compared with the TC+CC genotype group ($14 \pm 1$ vs $17 \pm 1$, $P = 0.09$).

We calculated the percent change in FBF and FVR from the baseline values at min 1, 2, and 3 during reactive hyperemia. There was a tendency for a main effect of eNOS genotype on both the percent change in FBF and FVR during peak vasodilation (Fig. 1). The TT genotype group had a greater percent increase in FBF than the TC+CC genotype group ($186 \pm 19$ vs $139 \pm 19\%$, $P = 0.05$), which corresponded to a greater percent decrease in FVR at min 1 ($-62 \pm 2$ vs $-51 \pm 4\%$, $P = 0.04$). There were no main effects of eNOS genotype with respect to any of the blood pressure variables.

**DISCUSSION**

The present study examined the potential interactive effects between the T-786C eNOS gene polymorphism and habitual physical activity level on FVR and FBF at rest and during reactive hyperemia in young Caucasian women who were participating in a larger study. We found significant interactive effects on resting FVR and FBF, and a significant main effect of eNOS genotype on peak FBF, and the percent change in FVR at peak vasodilation. Within the sedentary group, the TT genotype group had a lower resting FVR than the TC+CC group. However, within the endurance-trained group, the TC+CC and the TT genotype groups had similar resting FVR values. This finding was reflected in the resting FBF values. After accounting for
baseline FBF and percent body fat. Peak FBF was significantly greater, and the percent decrease in FVR at min 1 was significantly lower in TT genotype subjects. This agrees with the previous finding of reduced transcriptional and promoter activity in C allele carriers (25). Interestingly, by minute 3 of reactive hyperemia, FBF returned to resting levels only in the endurance-trained TT genotype group. This capacity to restore homeostasis more quickly may indicate the favorable response, and thus, the endurance-trained TT genotype group may have the best overall peripheral vascular function response.

Currently, there are no studies that have assessed the interactive effect of habitual physical activity level and genetics on FVR and FBF at rest and during reactive hyperemia. The sedentary TC+CC group clearly demonstrated the greatest resting FVR and the lowest FBF compared with any other group. Even the endurance-trained group with the TC+CC genotype had a significantly lower baseline FVR and significantly greater resting FBF than the sedentary TC+CC group. However, the sedentary subjects with the TT genotype and the endurance-trained subjects with the TT genotype had virtually the same baseline FVR values. These findings suggest that perhaps individuals can overcome the reported negative effects associated with having a C allele with chronic endurance exercise. These results also suggest that in subjects with the TT genotype, those reported to have “normal” vasomotor and cardiovascular function, cardiovascular fitness has less of an effect on resting FVR and FBF.

Aerobic exercise training reduces the risk of cardiovascular disease and all-cause mortality primarily by decreasing cardiovascular disease risk factors (2). It is thought that the endothelium is an important component of cardiovascular health (22). We found a significant main effect of physical activity level on resting FVR with the sedentary subjects demonstrating greater FVR than the endurance-trained subjects. However, this effect was primarily due to the sedentary C allele carriers who had the highest average resting FVR. This agrees with longitudinal studies that have shown improved endothelial function with aerobic exercise training (6,9,15). The physical activity groups clearly had different cardiovascular fitness levels as evidenced by the groups’ average VO_{2max} values. The sedentary group had an average VO_{2max} of 33.8 ± 1 mL·kg^{-1}·min^{-1}, which is in the 40th percentile for their gender and age (1). In contrast, the endurance-trained women, had an average VO_{2max} of 45.3 ± 1 mL·kg^{-1}·min^{-1}, which is in the 90th percentile (1).

The T-786C polymorphism has potentially important functional characteristics. The polymorphism is located in the promoter region (12,31) of the eNOS gene and therefore potentially affects gene expression levels. Nakayama et al. (25) found that individuals with a C allele had 50% decreased promoter activity compared with individuals without a C allele, which is consistent with the concept that the C allele is associated with reduced NO production. Interestingly, one study showed that a shear stress responsive element is located near the promoter region of the eNOS gene between nucleotides −1600 and −779 (18). Our results agree with these previous findings because the TT genotype individuals had a greater reduction in FVR and a greater increase in FBF than the C allele carriers during peak vasodilation. Our results also agree with a recent report by Rossi et al. (26), who found a blunted percent increase in FBF to incremental doses of acetylcholine in hyperventilative C allele carriers compared with the TT genotype individuals. However, Rossi et al. did not report on the physical activity status or body composition of their subjects, which can influence blood flow capacity.

In the present study, subjects were categorized into a combined TC+CC genotype group and compared with the TT genotype group. Based on available data, the frequency of the CC genotype in the population is between 0.02 and 0.10. Given these frequencies, we would have had to recruit between 150 and 750 subjects to fill the CC genotype group. In the present study, six subjects (10%) were CC homozygotes, which agrees with the reported frequency of the CC genotype. Thus, we did not have adequate statistical power to include a separate CC genotype group. Furthermore, the C allele is considered to have deleterious cardiovascular effects, with CC and TC genotype groups demonstrating similar responses that are different than groups with the TT genotype (25,26,34). Thus, it appears appropriate from a mechanistic perspective to group the C allele carriers into a combined TC+CC genotype group.

Ischemia is an extreme physiological condition and the response to ischemia is an increase in blood flow to protect the tissues against hypoperfusion (17). The extent to which NO is produced during postschemia vasodilation in the human forearm is debatable; however, several studies that used NO synthase inhibitors found modest reductions in peak reactive hyperemic blood flow and/or total hyperemic response (8–10,20). Our purpose was not to determine the role that NO plays in reactive hyperemia, but rather, given the functional characteristics of the T-786C polymorphism (12,24), we sought to determine whether there was a genotype-dependent difference in FVR and FBF. Our purpose was also not to determine whether the eNOS T-786C gene polymorphism was associated with endothelial function. This has been demonstrated by Rossi et al. using pharmacologic methods (26).

Our purpose was to determine the overall role of the eNOS T-786 gene polymorphism during a physiological stimulus that induces vasodilation. There are several other local regulators of blood flow, many of which interact with the NO system to produce postschemia vasodilation (4,11,13,21). Because eNOS and NO interact with other regulators of blood flow, it was important that we first assess the potential interactive effects of the eNOS gene and physical activity level on the global FVR and FBF response during reactive hyperemia. Other regulators of local blood flow that could potentially interact with eNOS to affect postschemia vasodilation are adenosine (3,27,32), prostaglandin (8,13,21,24,35), and adrenergic mechanisms (29,35). One study found that another eNOS gene polymorphism was associated with adenosine-induced hyperemia (23). Kilbom and Wennmalm (13) showed that prostag-
landin’s most prominent role was in postischemia and metabolic vasodilation in the human forearm and under certain metabolic conditions, prostaglandin modulates eNOS gene expression (24). Finally, studies have shown that NO counteracts sympathetic vasoconstrictor mechanisms (35) and is involved in the $\alpha_2$-adrenergic receptor-mediated endothelial-dependent vasodilation (30).

Multiple genes likely influence the reactive hyperemic response, with each gene contributing to a portion of the overall blood flow response. Thus, the influence of any single gene on the FVR and FBF response during reactive hyperemia is likely to be small. The eNOS T-786C gene polymorphism is a particularly attractive candidate because functional characteristics, namely, altered gene promoter activity and its proximity to a shear stress responsive element. Hypercholesterolemia impairs endothelial function and could thereby influence the reactive hyperemic response (7). The subjects in the present study were young, healthy, not obese, and none of them indicated the presence of any diseases or conditions that would impair endothelial function in their health history questionnaire. Therefore, it is unlikely that differences in cholesterol levels between the eNOS genotype groups contributed substantially to the differences in FVR and FBF observed in the present study. It is more likely that there were differences in cholesterol between the physical activity groups, but the only variable that was different between these groups was baseline FVR.

In conclusion, the present study found an interactive effect between physical activity level and the eNOS T-786C gene polymorphism on resting FVR and FBF. Among the sedentary subjects, those with the TT genotype and the lowest baseline FVR, but among the endurance-trained group, the TC+CC and TT genotype groups had identical baseline FVR values. With respect to FBF, sedentary subjects with the TT genotype had greater baseline FBF than the TC+CC genotype subjects; however, among the endurance-trained group, the TC+CC genotype group had a greater baseline FBF than the TT genotype subjects. Although the present investigation was a cross-sectional study, these results suggest that young female subjects possessing a C allele may reduce their resting FVR and increase their resting FBF by improving their cardiovascular fitness level. In contrast, young female sedentary TT homozygotes, who may have normal eNOS gene function, may not improve their resting FVR with improvements in cardiovascular fitness. Regardless of physical activity level, young females with the TT genotype had a significantly greater peak FBF compared with the C allele carriers. Most importantly, we found that after accounting for baseline FBF and percent body fat, peak FBF was significantly greater and the percent decrease in FVR at minute 1 was significantly lower in TT genotype subjects. These latter results agree with the report that the C allele is associated with reduced eNOS gene transcriptional and a promoter activity, which is consistent with the notion of reduced NO production.

We thank Gloria Sheng for her assistance in creating and managing the database.

REFERENCES


