**Supplementary Material S1**

**Table S1.** Genetic variants under study

|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **GENE** | **VARIANT** | **ALLELE** | **AA** | **GENE** | **VARIANT** | **ALLELE** | **AA** | **GENE** | **VARIANT** | **ALLELE** | **AA** |
| ***ABO*** | rs495828 | T>G | Upstream | ***BDKRB2*** | rs2227279 | G1061A | Gly354Glu | ***REN*** | rs5707 | A>C/G/T | Intron |
| ***ACE*** | rs2229839 | C1052T | Pro351Leu | ***CACNA1C*** | rs16929470 | C>T | Intron variant | ***REN*** | rs61746500 | G22C | Pro8Ala |
| ***ACE*** | rs3730025 | A765G | Tyr244Cys | ***CACNA1C*** | rs2239050 | G>A | Intron variant | ***REN*** | rs6704321 | C1207T(G) | Ala403Thr (Pro) |
| ***ACE*** | rs3730043 | C1025T | Thr342Met | ***CYP2D6*** | rs5030656 | CTT/- | Lys281Del | ***SCNN1A*** | rs13306616 | G1382T | Pro461His |
| ***ACE*** | rs4298 | C488T | Thr163Met | ***CYP3A4*** | rs2246709 | G>A | Intron | ***SCNN1A*** | rs5742912 | A1477G | Trp552Arg |
| ***ACE*** | rs4314 | C1681T | Arg561Trp | ***CYP3A4*** | rs2740574 | C>A/G/T | Upstream variant | ***SCNN1A*** | rs72657541 | T806C | Met628Thr |
| ***ACE*** | rs4364 | C2134A | Arg712Ser | ***CYP3A5\*3*** | rs776746 | T>C | intron | ***SCNN1A*** | rs72657550 | G1736C | Gly579Ala |
| ***ACE*** | rs4976 | T1331C | Ile444Thr | ***CYP3A5\*6*** | rs10264272 | G624A | Lys198= | ***SCNN1B*** | rs250563 | C>T | Phe293Phe |
| ***ACE*** | rs4291 | T>A | Upstream variant | ***CYPIIB2*** | rs1799998 | G>A | Upstream | ***SCNN1B*** | rs41278184 | C1136T | Thr379Met |
| ***ACE*** | rs4646992 | Indel | Intron variant | ***GNB3*** | rs5443 | C825T | Ser275Ser | ***SCNN1B*** | rs149868979 | C1706G>A | Arg563Gln |
| ***ADD1*** | rs4961 | G1378T | Gly460Trp | ***Intergenic*** | rs2820037 | A>T | Intergenic | ***SCNN1B*** | rs72654326 | C910T | Arg259Trp |
| ***ADRB2*** | rs1042714 | G>C | Gln27Glu | ***Intergenic*** | rs11646213 | A>T | Intergenic | ***SCNN1B*** | rs72654356 | T2018A | Leu628Gln |
| ***AGT*** | rs11122576 | A>T | Intron variant | ***KLK1*** | rs5518 | T578A | Val193Glu | ***SCNN1G*** | rs5723 | C19477G | Leu649Leu |
| ***AGT*** | rs4762 | G620A | Thr207Met | ***KNG1*** | rs4686799 | T>C | Intron | ***SCNN1G*** | rs5729 |  | 3'UTR |
| ***AGT*** | rs61751067 | G1261A | Val421Met | ***KNG1*** | rs5030062 | A>C | Non-coding transcript | ***SCNN1G*** | rs7200183 |  | Intron |
| ***AGT*** | rs61751077 | C1325T | Ser442Phe | ***KNG1*** | rs698078 | A>G | Intron variant | ***SCNN1G*** | rs72646501 | C776A | Thr259Asn |
| ***AGT*** | rs61762527 | C1144G | Pro382Ala | ***LDLR*** | rs688 | C1773T | Asn464Asn | ***SCNN1G*** | rs72647542 | C1868T | Pro623Leu |
| ***AGT*** | rs61762537 | C709T | Arg237Cys | ***LOC124900725*** | rs1458038 | C>T | intron | ***SLC12A3*** | rs2399594 | A>G | Intron |
| ***AGT*** | rs61762540 | T251C | Leu84Pro | ***LPL*** | rs328 | C1421G | Ser474NULL | ***SLC14A2*** | rs1123617 | G>A | Val750Ile |
| ***AGT*** | rs61762541 | G11C | Arg4Pro | ***NOS1AP*** | RS10494366 | G>C/T | Intron | ***SLC14A2*** | rs3745009 | G2638A | Ala880Thr |
| ***AGT*** | rs699 | A8030G | Met268Thr | ***NOS3*** | rs1799983 | T894G | Asp298Glu | ***SLCO1B1*** | rs4149056 | T521A | Val174Glu |
| ***AGT*** | rs3889728 | G>C | Intron | ***NOS3*** | rs2070744 | C>T | Intron | ***TGFB1*** | rs1800471 | G74C | Arg25Pro |
| ***AGTR1*** | rs12721225 | G817T | Ala273Ser | ***NOS3*** | rs41508746 | C1114T | Arg372Cys | ***TNFRSF1A*** | rs4149570 | A>C | Upstream |
| ***AGTR1*** | rs13095608 | T209G | Val70Gly | ***NOS3*** | rs61747096 | C2831T | Ser944Leu | ***TRPC7*** | rs2277052 | G>A | Intron |
| ***AGTR1*** | rs5182 | C573T | Leu220Leu | ***NPPA-AS1*** | rs5065 | A454G | NULL152Arg | ***TRPC7*** | rs2277052 | G>A | Intron |
| ***AGTR1*** | rs5186 | A>C | 3' UTR | ***NR2F2-AS1*** | rs2398162 | A>G | Non-coding transcript | ***WNK1*** | rs2107614 | T>A | Intron |
| ***AGTR2*** | rs3729979 | C812T | Pro271Leu | ***NR3C2*** | rs5523 | A1331C | Asn444Thr | ***WNK1*** | rs2286007 | C1994T | Thr665Ile |
| ***ATP2B1*** | rs17249754 | C>T | Intron variant | ***NR3C2*** | rs5522 | G538A | Val108Phe | ***WNK1*** | rs2277869 |  | intron |
| ***BDKBR2*** | rs11847625 | G>A/C | Intron | ***PTGIS*** | rs5629 | G938T | Arg373Ag | ***YEATS4*** | rs315135 | A>G | Intron |
| ***BDKRB2*** | rs1046248 | C>T | Arg14Cys | ***REN*** | rs11571098 | G>A | Arg33Trp | ***YEATS4*** | rs7297610 |  | Intergenic |
| ***BDKRB2*** | rs1799722 | C>T | 5' UTR | ***REN*** | rs2368564 | C>T | Intron | Intergenic | rs13278559 | C>T | 5' UTR |

**Supplementary Material S2**

The ACE insertion-deletion was genotyped as previously described (49 – 51}. Briefly, all reactions were made up to a final volume of 25μl. A standard reaction contained 100ng of template DNA, 1.5 units (U) of GoTaq DNA polymerase (Promega, USA), 1 x GoTaq Polymerase Buffer (pH 8.5, 1.5mM MgCl2 per reaction), a final concentration of 200 micromolar (μM) deoxyribonucleotide (dNTPs) (Bioline, USA), 5% Dimethyl sulfoxide (DMSO) (Merck, USA), 50mM KCl (Merck, USA), 10mM Tris-HCL (Merck, USA), 0.1% Triton X-100 (Merck, USA ), a final concentration of 10pM of each primer (forward and reverse) and distilled water (Sabex) made up to the final volume.

**Table S2a**. *ACE* Insertion-Deletion Primer pairs [49 - 51]

|  |  |
| --- | --- |
| **Primer Name** | **Primer Sequence** |
| Forward Primer | 5’ CTGGAGAGCCACTCCCATCCTTTCT 3’ |
| Reverse Primer | 5` GATGTGGCCATCACATTCGTCAGAT 3` |
| Forward Insertion Specific Primer | 5` TGGGACCACAGCGCCCGCCACTAC 3` |
| Reverse Insertion Specific Primer | 5` TCGCCAGCCCTCCCATGCCCATAA 3` |

The standard cycling conditions used for the PCR included: an initial denaturing step at 94° C for 5 minutes, 30 cycles of amplification, which involved a denaturing step at 94° C for 30 seconds; an annealing temperature of 67° C for 60 seconds and an elongation step at 72°C for 2 minutes. A final step of 72°C for 5 minutes ensured completion of the reaction. PCR products were detected by Agarose Gel Electrophoresis (AGE). The GeneRuler 100 base-pair (bp) molecular weight marker (Fermentas, USA) was employed for all resolution of fragments on an agarose gel. The marker was used at a concentration of 0.05 micrograms (μg)/μl. The PCR products were then analysed manually to determine the size of the product and genotyped accordingly. Insertion homozygotes presented with one band at 490 base pairs while insertion heterozygotes presented with one band at 490 base pairs and another at 190 base pairs. Deletion homozygotes only presented with a single band at 190 base pairs. The insertion specific PCR yielded a product of 335bp in the presence of te insertion, while no product was observed in homozygous deletion samples.

**Supplementary Material S3**

|  |  |  |  |
| --- | --- | --- | --- |
| Study Population | Female | Male | Total |
| Hypertensive Individuals | 163 | 114 | 277 |
| Normotensive Individuals | 114 | 62 | 176 |
| Total | 277 | 176 | 453 |

**Table S3a.** Stratification of the study population by sex

**Supplementary Material S4**

**Table S4a.** Variants significantly associated with EH in the female cohort under study.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **GENE** | **SNP ID** | **Genotype** | **Female** | | | | |
| **Hypertensive (N =163)** | **Hypertensive (%)** | **Normotensive (N = 114)** | **Normotensive (%)** | **p-Value** |
| ***NOS3*** | **rs1799983** G894T | GG | 11 | 6.75% | 18 | 15.79% |  |
| GT | 28 | 17.18% | 40 | 35.09% | 3.84e-05 |
| TT | 120 | 73.62% | 56 | 49.12% |  |
| ***CYP11B2*** | **rs1799998**  -344C>T | CC | 89 | 54.60% | 20 | 17.54% | 1.19e-15 |
| CT | 62 | 38.04% | 42 | 36.84% |
| TT | 12 | 7.36% | 52 | 45.61% |
| ***AGT*** | **rs5051**  -30-3273G>T | GG | 11 | 6.75% | 50 | 43.86% | 0.00011 |
| GT | 72 | 44.17% | 36 | 31.58% |
| TT | 80 | 49.08% | 28 | 24.56% |
| ***AGTR1*** | **rs5186** A1166C | AA | 54 | 33.13% | 80 | 70.18% | < 2.2e-16 |
| AC | 27 | 16.56% | 28 | 24.56% |
| CC | 82 | 50.31% | 6 | 5.26% |
| ***AGT*** | **rs699**  T776C | TT | 6 | 3.68% | 18 | 15.79% | 5.79e-06 |
| TC | 39 | 23.93% | 44 | 38.60% |
| CC | 118 | 72.39% | 52 | 45.61% |
| ***ACE*** | **rs4646994**  INDEL | II | 33 | 20.25% | 22 | 19.30% | 1.00e-06 |
| ID | 75 | 46.01% | 82 | 71.93% |
| DD | 55 | 33.74% | 10 | 8.77% |

**Table S4b.** Variants significantly associated with EH in the male cohort under study.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **GENE** | **SNP ID** | **Genotype** | **Male** | | | | |
| **Hypertensive (N =110)** | **Hypertensive (%)** | **Normotensive (N = 62)** | **Normotensive (%)** | **p-Value** |
| ***CYP11B2*** | **rs1799998**  -344C>T | CC | 32 | 29.09% | 6 | 9.68% | 4.40e-09 |
| CT | 45 | 40.91% | 18 | 29.03% |
| TT | 13 | 11.82% | 38 | 61.29% |
| ***AGT*** | **rs5051**  -30-3273G>T | GG | 23 | 20.91% | 36 | 58.06% | 4.72e-05 |
| GT | 62 | 56.36% | 20 | 32.26% |
| TT | 13 | 11.82% | 6 | 9.68% |
| ***AGTR1*** | **rs5186** A1166C | AA | 0 | 0.00% | 44 | 70.97% | < 2.2e-16 |
| AC | 29 | 26.36% | 16 | 25.81% |
| CC | 61 | 55.45% | 2 | 3.23% |
| ***ACE*** | **rs4646994**  INDEL | II | 13 | 11.82% | 16 | 25.81% | 1.39e-05 |
| ID | 46 | 41.82% | 38 | 61.29% |
| DD | 51 | 46.36% | 8 | 12.90% |

**Supplementary Material S4**

**Table S4a.** Alleles significantly associated with EH in the female cohort

|  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  |  |  |  |  |  | **Female** |  |  |  |
| **Gene** | **SNP ID** | **Allele** | **Hypertensive (N =326)** | **Hypertensive (%)** | **Normotensive**  **N = 228)** | **Normotensive (%)** | **p-Value** | **95% CI** | **OR** |
|  |
| **CYP11B2** | **rs1799998** | C | 201 | 61.66% | 82 | 35.97% | 2.49e-0.6 | 1.620 - 3.412% | 2.35 |  |
| -344C>T | T | 125 | 38.34% | 146 | 64.03% |  |
| **Intergenic** | **rs2820037** | A | 243 | 74.54% | 46 | 20.18% | 4.24e-05 | 1.522 - 3.542% | 2.30 |  |
| Intergenic | T | 83 | 25.46% | 182 | 79.82% |  |

**Table S4b.** Alleles significantly associated with EH in the male cohort

|  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
|  |  |  | **Male** | | | | | | |
| **Gene** | **SNP ID** | **Allele** | **Hypertensive**  **(N =220)** | **Hypertensive (%)** | **Normotensive (N =124)** | **Normotensive (%)** | **p=Value** | **95% CI** | **OR** |
|  |
| **CYP11B2** | **rs1799998** | C | 157 | 71.36% | 30 | 24.19% | 5.67e-16 | 4.104 - 11.80 | 6.89 |  |
| -344C>T | T | 71 | 32.27% | 94 | 75.81% |  |
| **AGTR1** | **rs5186** | A | 77 | 35,00% | 104 | 83.87% | <2.2e-16 | 0.0536 - 0.1747 | 0.0987 |  |
| A1166C | C | 143 | 65,00% | 20 | 16.13% |  |
| **ACE** | **rs4646994** | I | 72 | 32.72% | 70 | 56.45% | 0.0001247 | 0.2600 - 0.6683 | 0.4180 |  |
| INDEL | D | 148 | 67.27% | 54 | 43.55% |  |