**Table 1. Summary on the ovarian tumor samples (N = 122)**

|  |  |
| --- | --- |
| **Clinical and pathological parameters** | **Absolute numbers** |
| Type of ovarian tumor: |  |
| Borderline tumor | 18 |
| Malignancy | 104 |
| Histological type: |  |
| Serous adenocarcinoma | 85 |
| Endometrioid adenocarcinoma | 15 |
| Others | 22 |
| Stage: |  |
| I | 26 |
| II | 19 |
| III | 68 |
| IV | 9 |
| Tumor size and extension/spread: |  |
| Т1 | 26 |
| Т2 | 21 |
| Т3 | 75 |
| Differentiation grade: |  |
| G1 | 25 |
| G2 | 31 |
| G3 | 48 |
| Hematogenous metastasis: |  |
| M0 | 113 |
| M1 | 9 |
| Lymphogenous metastasis: |  |
| N0 | 101 |
| N1–3 | 21 |
| Peritoneal dissemination: |  |
| No | 77 |
| Yes | 45 |
| Omental metastasis: |  |
| No | 61 |
| Yes | 61 |
| All types of metastasis: |  |
| No | 43 |
| Yes | 79 |
| Ascitis: |  |
| No | 53 |
| Yes | 49 |

**Table 2. Primers and conditions for methyl-specific polymerase chain reaction used in the study**

|  |  |  |
| --- | --- | --- |
| **lncRNA gene / primer, nucleotide sequence** | **Length, bp** | **Annealing temperature, ºС** |
| *GAS5* |  |  |
| MF: CGTTATCGTCGGTATTGGAGGGG | 185 | 60 |
| MR: CGCCCGACGCCTTATCCC |
| UF: TGTTATTGTTGGTATTGGAGGGGTGAG | 179 | 60 |
| UR: CAACACCTTATCCCCATCTTCTCCA |
| *HOTAIR* |  |  |
| MF: CGGGTTTTTATTTTTTCGTTATTGCG | 258 | 54 |
| MR: CGACTACTCTCGCCAAATTTCACTACTTC |
| UF: TGGGTTTTTATTTTTTTGTTATTGTGTTATTTTG | 258 | 52 |
| UR: CAACTACTCTCACCAAATTTCACTACTTCACAC |
| *LINC00472* |  |  |
| MF: AAGGCGTTTTAAGTCGAGGGTA | 224 | 60 |
| MR: AACGACTCCGACAACACACC |
| UF: AAGGTGTTTTAAGTTGAGGGTAAAG | 228 | 59 |
| UR: AACAACTCCAACAACACACCCAC |
| *LINC00886* |  |  |
| MF: CGTGCGATCGTAGTTCGGTAGGTTA | 172 | 60 |
| MR: CGCCGAATTACGCGACGAAA |
| UF: CGTGCGATCGTAGTTCGGTAGGTTA | 181 | 60 |
| UR: CCTCACCAAATTACACAACAAAATCAACAC |
| *MAFG-DT* |  |  |
| MF: CGGATTTTCGGGCGTTTCG | 232 | 60 |
| MR: ATTTCGAATCTACCGCGCAC |
| UF: TGTGGATTTTTGGGTGTTTTGTTTG | 236 | 60 |
| UR: ATTTCAAATCTACCACACACCC |
| *PLUT* |  |  |
| MF: CGGGGATTTGGTATTGTGTGGC | 201 | 60 |
| MR: CTAAACCTAACCTCTTAATACGACCAACCA |
| UF: TGTTGGAATGTGTATGGGTTTTTGTAAAGTT | 339 | 61 |
| UR: CACAAATACCTAAACCTAACCTCTTAATACAACCA |
| *SNHG1* |  |  |
| MF: CGGCGATCGAGGTTTTAGGA | 210 | 60 |
| MR: ACTAACTCACCGACCGCATT |
| UF: TGGTGATTGAGGTTTTAGGA | 210 | 55 |
| UR: ACTAACTCACCAACCACATT |
| *SNHG6* |  |  |
| MF: TTGAGTTATCGCGTTCGGTTT | 295 | 61 |
| MR: CTCTTCCGATACGCGACCC |
| UF: CTCTTCCAATACACAACCC | 295 | 58 |
| UR: CAAAAACCATAAACCACCCTCC |
| *SNHG12* |  |  |
| MF: CGCGTTTAGTAAAATTATATATTAGTGGAAGAGATAAG | 239 | 60 |
| MR: CCCGACGCTAAACCCACGC |
| UF: TGTGTTTAGTAAAATTATATATTAGTGGAAGAGATAAG | 245 | 56 |
| UR: TCAATACCCAACACTAAACCCACAC |
| *SNHG17* |  |  |
| MF: GCGCGAAACGAGCGTA | 168 | 59 |
| MR: CGACGCCCTAACGTCGAATA |
| UF: TTGGTGTGAAATGAGTGTA | 170 | 57 |
| UR: CAACACCCTAACATCAAATAACA |
| *TINCR* |  |  |
| MF: GCGGACGAGGCGTTGTTGTTAT | 193 | 60 |
| MR: CGCTAACGAACAACAACACCGAAC |
| UF: GTGGATGAGGTGTTGTTGTTATTGTTGATT | 194 | 60 |
| UR: TCACTAACAAACAACAACACCAAACCATC |
| *TP53TG1* |  |  |
| MF: TCGTTTCGTGTTTGACGTC | 137 | 55 |
| MR: ACTCATTTAACACCCGACGA |
| UF: GTTTTGTTTTGTGTTTGATGTT | 137 | 55 |
| UR: ACTCATTTAACACCCAACAAACC |
| *TUG1* |  |  |
| MF: CGGGTTTCGGTTTCGTGGTC | 199 | 60 |
| MR: CGACGAAAACGACAACAACACATAATT |
| UF: TGGTTTTTAAGGATTGGATTGAGGGTAG | 159 | 60 |
| UR: CAACAACAACAAAAACAACAACAACACATAAT |

MF/UF and MR/UR, direct and reverse primers to the methylated/ non-methylated allele, respectively. Oligonucleotides were selected with https://www.ncbi.nlm.nih.gov/gene/ database and http://www.urogene.org/methprimer2/ software, controlled by SeqBuilder Pro software, which is a part of the Lasergene 17.1 software package (DNASTAR, USA). The oligonucleotides for the *TP53TG1* gene were taken from [19]

**Table 3. Methylation levels for two lncRNA genes in the tumors of the patients with and without lymphatic node metastasis**

|  |  |  |  |
| --- | --- | --- | --- |
| **lncRNA genes** | **Lymphogenous metastasis** | **Methylation level, %** | **P value** |
| *SNHG6* | N0 | 24.41 [12.14; 47.51] | 0.044 |
| N1–3 | 50.59 [12.12; 64.11] |
| *SNHG12* | N0 | 21.42 [7.49; 37.34] | 0.006 |
| N1–3 | 42.95 [37.38; 48.84] |

Twenty one (21) samples from the patients with lymphatic node metastases and 101 samples from the patients without lymphogenous metastasis were studied; the values are given as medians (Me) and quartiles [Q1; Q3]

**Table 4. Methylation levels for four lncRNA genes in the tumors of the patients with and without peritoneal dissemination**

|  |  |  |  |
| --- | --- | --- | --- |
| **lncRNA genes** | **Peritoneal dissemination** | **Methylation level, %** | **P value** |
| *HOTAIR* | No | 4.49 [2.94; 5.64] | 0.001 |
| Yes | 6.39 [4.97; 7.83] |
| *TINCR* | No | 27.16 [5.86; 47.55] | 0.001 |
| Yes | 45.41 [37.69; 53.64] |
| *GAS5* | No | 4.46 [0.71; 9.68] | 0.026 |
| Yes | 8.60 [6.72; 9.60] |
| *LINC00472* | No | 2.14 [0.88; 5.44] | 0.041 |
| Yes | 5.20 [0.89; 8.30] |

Forty five (45) samples from the patients with peritoneal dissemination and 77 samples from the patients without peritoneal dissemination were studied; the values are given as medians (Me) and quartiles [Q1; Q3]

**Table 5. Methylation levels for four lncRNA genes in the tumors of the patients with and without omental metastasis**

|  |  |  |  |
| --- | --- | --- | --- |
| **lncRNA genes** | **Great omentum metastasis** | **Methylation level, %** | **P value** |
| *HOTAIR* | No | 4.49 [3.06; 5.77] | 0.003 |
| Yes | 6.30 [4.78; 7.83] |
| *GAS5* | No | 4.92 [0.77; 9.04] | 0.033 |
| Yes | 8.94 [7.05; 9.53] |
| *LINC00472* | No | 1.52 [0.75; 4.24] | < 0.001 |
| Yes | 6.42 [2.77; 8.25] |
| *LINC00886* | No | 6.16 [0.84; 13.57] | 0.021 |
| Yes | 9.62 [7.83; 12.86] |

Sixty one (61) samples from the patients with omental metastasis and 61 samples from the patients without omental metastasis were studied; values are given as medians (Me) and quartiles [Q1; Q3]

**Table 6. Methylation levels for eight lncRNA genes in the tumors of the patients with and without any type of metastasis, including distant**

|  |  |  |  |
| --- | --- | --- | --- |
| **lncRNA genes** | **Any type of metastasis** | **Methylation level, %** | **P value** |
| *HOTAIR* | No | 4.38 [3.01; 5.45] | 0.002 |
| Yes | 6.05 [3.73; 7.57] |
| *TUG1* | No | 4.66 [0.71; 5.52] | 0.024 |
| Yes | 5.455 [3.8; 6.48] |
| *SNHG6* | No | 16.95 [8.96; 35.08] | 0.005 |
| Yes | 35.44 [12.73; 60.29] |
| *TINCR* | No | 22.84 [6.05; 47.87] | 0.039 |
| Yes | 44.18 [25.0; 49.27] |
| *GAS5* | No | 3.3 [0.57; 8.97] | 0.022 |
| Yes | 8.44 [2.08; 9.64] |
| *LINC00472* | No | 1.4 [0.71; 2.78] | < 0.001 |
| Yes | 6.095 [1.375; 8.26] |
| *LINC00886* | No | 4.49 [0.83; 14.5] | 0.046 |
| Yes | 9.5 [6.34; 12.84] |
| *MAFG-DT* | No | 5.53 [1.93; 7.42] | 0.039 |
| Yes | 2.355 [1.22; 6.13] |

Seventy nine (79) tumor samples from the patients with any metastasis 43 samples from the patients without metastasis were studied; values are given as medians (Me) and quartiles [Q1; Q3]

**Table 7. Comparison of methylation levels of four lncRNA genes in macroscopic peritoneal metastasis and their primary tumors**

|  |  |  |  |
| --- | --- | --- | --- |
| **lncRNA gene** | **Tumor / macroscopic metastasis** | **Methylation level, %** | **P value** |
| *MAFG-DT* | Tumor | 2.41 [1.26; 4.96] | < 0.001 |
| Macro metastasis | 6.2 [5.38; 8.11] |
| *TP53TG1* | Tumor | 2.85 [1.38; 5.46] | < 0.001 |
| Macro metastasis | 6.2 [4.56; 8.18] |
| *SNHG12* | Tumor | 9.16 [7.67; 23.82] | 0.002 |
| Macro metastasis | 1.88 [0.23; 18.26] |
| *LINC00886* | Tumor | 8.47 [6.38; 12.76] | 0.003 |
| Macro metastasis | 6.44 [3.82; 8.27] |

Forty five (45) samples of macroscopic peritoneal metastases related to 45 samples of their primary tumors were studied; the values are given as median (Me) and quartiles [Q1; Q3]

**Table 8. Methylation levels for two lncRNA genes in the tumors of the patients with and without ascitis**

|  |  |  |  |
| --- | --- | --- | --- |
| **lncRNA genes** | **Ascitis** | **Methylation level, %** | **P value** |
| *LINC00472* | No | 1.99 [0.71; 6.09] | 0.022 |
| Yes | 5.80 [0.89; 8.51] |
| *LINC00886* | No | 5.34 [0.83; 9.49] | 0.022 |
| Yes | 9.04 [4.96; 12.75] |

Forty nine (49) tumor samples from the patients with ascites and 53 tumor samples from the patients without ascites were studied; the values are given as median (Me) and quartiles [Q1; Q3]