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Identification of miRNAs Associated with Osteoblastic Differentiation

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At 21-25 nucleotides long, MicroRNAs (miRNA) are non-coding, regulators of gene expression at the posttranscriptional level through degradation and/or inhibition of mRNA translation. miRNAs have been associated with various human diseases, although its precise role in metabolic bone diseases such as osteoporosis is unclear. The aim of this study is to investigate the involvement of miRNA at different stages of human bone development. We investigated miRNA expression in three human osteosarcoma cell lines representing different stages of osteoblast differentiation, MG-63 the least differentiated, TE-85 intermediate and SaOS-2 the most mature. In the other study, we found that SaOS-2 expressed the highest level of sclerostin and alkaline phosphatase followed by TE85 and MG63. This result confirms that SaOS-2 is the most mature while MG63 is the least differentiated. All cell lines were cultured in DMEM + 10% FCS. Total RNA was extracted from confluent cell culture. More than 500 miRNAs were differentially expressed, those with the largest difference in expression were hsa-miR-935, hsa-miR-143-3p, hsa-miR-145-5p, hsa-miR-155-5p, hsa-miR-3200-3p, hsa-miR-584-5p, hsa-miR-486-3p, hsa-miR-767-5p and hsa-miR-105-5p. miR-935 was expressed highest in TE85 and lowest in SaOS-2 ($p<0.05$). Expression of miR-143-3p and miR-155-5p was significantly higher in TE85 and SaOS-2 cells compared to MG63 ($p<0.05$). However, MG63 showed highest expression of miR-155-5p, miR-3200-3p and miR-584-5p compared with TE85 and SaOs-2 ($p<0.05$). The most striking observation to emerge from the data comparison was the expression of miR-155-5p in MG63 was 2842-fold greater than SaoS-2 and 1467-fold greater in TE85 than SaOS-2. Meanwhile, miR-486-3p, miR-767-5p and miR-105-5p were highest in SaOS-2 compared with MG63 and TE85 ($p<0.05$). Our data shows that different stages of osteoblast development are characterised by different sets of highly expressed microRNAs and suggests that miRNAs could be potential biomarkers in bone development and may provide the basis of new therapeutic approaches to prevent bone loss.

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Mutations in the *LRP4* and *LRP5* Genes are Associated with Bone Mineral Density in Maltese Postmenopausal Women

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Background: Osteoporosis is a multifactorial skeletal disease characterised by low bone mass leading to increased fracture risk. Members of the low-density lipoprotein receptor-related protein (*LRP*) gene family play a role in osteoblastogenesis through the Wingless (Wnt)/βeta-catenin pathway. *LRP4* controls the actions of sclerostin, a Wnt inhibitor, whereas *LRP5* promotes bone formation. The aim was to evaluate the effect of four non-synonymous coding polymorphisms in relation to bone mineral density (BMD) and low-trauma fractures in Maltese postmenopausal women. Genotyped variants were the *LRP4* rs2306033 (C>T) and rs6485702 (G>A) variants, and the *LRP5* rs4988321 (G>A) and rs3736228 (C>T) variants.
Methods: Research subjects were 1045 women aged 40 to 79 years, subdivided in three BMD groups without a fracture history: normal, osteopenic or osteoporotic. Women with a fracture history were classified as cases. Genotyping was performed by polymerase chain reaction and restriction fragment length polymorphism, or by real-time PCR. Odds ratios were computed using logistic regression analysis adjusted for age and clinical risk factors.
Results: Homozygosity for the rs6485702 G allele was found associated with low BMD at the lumbar spine, LS (P=0.01) relative to research subjects with a normal BMD, whereas heterozygotes for this allele had a low BMD at the femoral neck, FN (P=0.04). Women carrying one or two copies of the rs3736228 T allele and one copy of the rs4988321 A allele had a lower BMD at the LS and FN (P<0.05). The rs6485702 and rs3736228 variants were associated with increased fracture risk, however this was not independent of BMD. Interactions were observed between these three variants at the LS (P<0.01). Women carrying the *LRP4* C-G haplotype and *LRP5* A-T haplotype had lower BMD measurements at the LS and FN (P<0.05).
Conclusion: The *LRP4* rs6485702 variant and the two *LRP5* variants play a role in BMD regulation in Maltese postmenopausal women.
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Polymorphisms in Neuropeptides Genes and Bone Mineral Density in Postmenopausal Korean Women

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Background: The purpose of this study was to investigate the association between single nucleotide polymorphisms in neuropeptide genes and bone mineral density (BMD) in postmenopausal Korean women.