

OSTA BIOTECHNOLOGIES INC.

OSTEOPOROSIS PROGNOSTIC TEST

Osta is well advanced in the development of a pre-onset genetics-based prognostic test for osteoporosis based on polymorphisms in the VNTR region of the human PTHrP gene. Osta's prognostic test is used for assessing the risk of developing osteoporosis and has a large market potential, given treatment costs and aging populations. The cost of treating fractures (surgery, implants, hospital and home care) and subsequent morbidity and mortality contributes significantly to the economic burden of osteoporosis. For this reason, the FDA and other government agencies, including public health care systems and HMOs, are receptive to novel diagnostic methods and prophylactic therapies for bone diseases, such as those being developed by Osta. Currently, there are no prognostic tests commercially available that can predict the risk of developing osteoporosis prior to the onset of the disease.

Osteoporosis is a highly prevalent skeletal disorder characterized by compromised bone strength that predisposes to an increased risk for fracture at the hip, spine, & other skeletal sites with significant associated morbidity and mortality. Osteoporosis and related fractures are common in aging individuals and contribute substantially to poor quality of life for those affected as well as increased healthcare costs and burden of illness to society at large. Although a number of factors such as rate of bone loss, bone size and structure, and propensity to fall contribute to the risk of osteoporosis fractures, the most important parameter in determining the risk for developing osteoporosis and ensuing fractures is the peak bone mass attained during young adulthood. Skeletal bone mass reaches over 90% of its maximum by age 18 (earlier in females than males). Peak bone mass is influenced by a number of environmental factors such as lifestyle, concomitant diseases, and diet. In addition, twin studies have suggested that up to 85% of the variance in bone mineral density is genetically determined depending on the site examined. Hence, genetically determined factors cause hereditary low bone mass, which reduces peak bone mass acquisition. Consequently bone mass is lower than the reference range at skeletal maturity (age 25-30 years). Patients with hereditary low bone mass, therefore, have less bone to lose in later life before fracture risk further increases and have low bone mineral density (BMD) early in adult life. Genetic factors may also increase bone loss in later life. It is highly desirable to identify those individuals who are at high risk for osteoporosis prior to the onset of the disease so that they can benefit from prophylactic interventions and earlier treatment.

Our researchers have shown earlier that Parathyroid hormone-related peptide (*PTHrP*) is expressed in the bone-forming cells of the skeleton (osteoblasts) and is critical for their proper proliferation, differentiation, and function, processes that are pivotal for attaining and maintaining appropriate bone mass thereby preventing the development of osteoporosis. Mice homozygous for *PTHrP* gene inactivation were shown to exhibit chondrodysplastic abnormalities and altered endochondral bone formation, that culminate to their death in the immediate peripartum period. On the other hand, mice heterozygous for the inactivated *PTHrP* allele are phenotypically normal at birth but develop by three

months of age premature and advanced osteoporosis. Moreover, mice generated with osteoblast-specific deletion of *PTHRP*, demonstrate impaired recruitment of osteoprogenitor cells and premature programmed cell death of osteogenic cells resulting in decreased bone formation and thereby, premature osteoporosis.

The human *PTHRP* gene (on chromosome region 12p12.1-p11.2) has also been reported to play an important role in development and specifically in skeletogenesis. Whether *PTHRP* expression within the human skeletal microenvironment is critical for attainment of peak bone mass during adolescence and maintenance of skeletal homeostasis later in adult life as in mice remains to be determined. Polymorphisms in the *PTHRP* gene have not been reported in association with either peak bone mass attained or propensity for osteoporosis and associated fracture risk. A variable number of tandem repeats (VNTR) polymorphism in *PTHRP* located in intron V, 100-bp downstream of exon VI has been reported. In the general population, it is characterized by eight different alleles ranging in length from 252 bp to 460 bp containing the repeat unit $[G(TA)nC]_N$, where $n = 4-11$ and $N = 3-17$.

Pilot human studies on 19 osteoporotic males conducted at a hospital in Montreal have provided an initial clinical proof of principal. Osta has initiated a second larger human clinical study on 80 healthy and osteoporotic male and female subjects at a hospital in Montreal. In addition, Osta has established a clinical research collaboration agreement with Helsinki University in Finland. The results of the collaborative research are expected to be available shortly.

In summary, our findings to date indicate that particular alleles and combinations of specific *PTHRP* VNTR alleles correlate with variability in bone mass and raise the possibility that this genotypic approach may serve as an important tool in identifying individuals with a genetic predisposition to low bone mass acquisition at a relatively young age and hence a higher risk for developing osteopenia and osteoporosis in later life.