non-diabetic patients, though diabetic patients had higher BMD. The same observation was made when FRAX was adjusted by model 2 (p=0.001) or by model 3 (p=0.001). HbA1c correlated inversely with FRAX adjusted with all three models.

Discussion

FRAX calculator does not include T2DM among secondary causes of OP and this precludes a proper risk assessment independent of BMD. Trabecular bone assessment (TBS) captures a larger portion of the diabetes-associated fracture risk than BMD, however TBS it is not fully independent of the BMD. We examined 3 models of adjusted FRAX in T2DM patients that showed an important increase in fracture risk prediction when adding BMD - independent risk factors into FRAX calculator.

Conclusion

T2DM patients have a greater risk of major osteoporotic fracture in 10 years at the same BMD compared with non-diabetic population. New models of FRAX adjusted for T2DM are needed in assessing the intervention threshold for OP/osteopenia of patients with T2DM.

Reproductive Endocrinology
CLINICAL STUDIES IN FEMALE REPRODUCTION I

SSRI Use in the Peripartum Period Regulates Mammary Gland Parathyroid Hormone Related Protein (PTHrP) by a Serotonylation-Dependent Mechanism

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SAT-009

During lactation, a woman experiences a considerable amount of bone loss and recent studies suggest bone deficits persist years postpartum. Furthermore, selective serotonin uptake inhibitors (SSRIs), which are often prescribed to women experiencing peripartum depression, have been linked to osteopenia. Serotonin signaling can increase parathyroid hormone related protein (PTHrP), a bone remodeling protein which liberates calcium for the milk. Additionally, fluoxetine (a common SSRI) results in increased mammary gland serotonin content and PTHrP, and treatment during the peripartal period reduced maternal bone mineral density. One proposed mechanism of serotonin action is by its covalent addition to proteins by transglutaminase (TG2), termed serotonylation. We therefore investigated whether the combination of fluoxetine and lactation can exacerbate maternal bone loss and the underlying mechanism. We hypothesized that SSRI-induced serotonin signaling in the lactating mammary gland increases PTHrP through a serotonylation-dependent mechanism. Treatment of mouse mammary epithelial cells (HC11) with fluoxetine significantly upregulates PTHrP gene expression and the concentration of its downstream effector, cAMP, over control ($P < 0.0004$). Furthermore, treatment of the HC11 cells with fluoxetine in addition to a TG2 inhibitor, monodansylcadaverine, restores PTHrP mRNA expression to levels observed in the control. Small g-proteins have emerged as a common target protein for serotonylation. Currently, our data suggest that the g-proteins, RhoA and Rab4, are potential serotonylation targets in the mammary gland. Together these data suggest that the molecular process of serotonyation in HC11 cells links serotonin signaling to increased PTHrP expression. Future work is directed at using the cre-lox system to genetically ablate serotonylation using a WAP^Cre/TG2^Flox transgenic mouse to determine whether decreasing serotonylation *in vivo* in the mammary gland during lactation improves maternal bone mass.

Tumor Biology
TUMOR BIOLOGY: DIAGNOSTICS, THERAPIES, ENDOCRINE NEOPLASIAS, AND HORMONE DEPENDENT TUMORS

The Roles of Two Insulin Receptor Isoforms in Triple Negative Breast Cancer Growth

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SUN-131

Systemic hyperinsulinemia is believed to be an important factor in the progression of a number of cancers, including breast cancer by activating the insulin receptor (IR) signaling cascade in the tumor cells. The IR is expressed in two isoforms, IR-A and IR-B. IR-B is the full-length isoform, while IR-A is lacking 12 amino acids in the α-subunit due to exon 11 alternative splicing. IR-A is predominantly expressed in cancer tissues, while IR-B is mostly expressed in metabolic tissues. The IR and closely related insulin-like growth factor 1 receptor (IGF-1R) are expressed in different ratios in cancer cells. Compared with estrogen receptor positive breast cancers, triple negative breast cancers (TNBC) frequently have higher ratios of IR to IGF-1R. Hyperinsulinemia is associated with increased prevalence of TNBC in pre-menopausal women. Although new targeted therapies are emerging, among breast cancer subtypes TNBC continues to carry the worst prognosis and therefore developing a greater understanding of the links between IR signaling and TNBC progression is critical. The aim of this study is to understand the role of IR-A and IR-B on proliferation, metastasis and metabolism in breast cancer cells. We stably overexpressed human IR-A (IR-A OE) and IR-B (IR-B OE) in TNBC MDA-MB-231 (231) and murine c-myc/vegf overexpressing Mvt1 cells with lentiviral transduction using pLVX-IRES-puro HIV-1-based expression vectors with cDNA encoding the human IR-A,IR-B and control cDNA sequences. Native murine IR was silenced using lentiviral transduction of shRNA in the Mvt1 cells. Overexpression of IR was confirmed at a protein level by western blot, and RNA isoform expression was confirmed using real time PCR. Cell proliferation assays were performed in DMEM/10% FBS and revealed that MDA-MB-231 cells with IR-A OE cells had 15% higher proliferation rates than 231 IR-B OE cells. We then examined the IR signaling pathways by western blot in DMEM/10% FBS. No differences in phosphorylated or total ERK1/2

were observed between control, 231 IR-A OE and 231 IR-B OE cells. 231 IR-A OE cells were found to have 15-fold greater Akt phosphorylation (Ser473) than 231 control cells (p=0.0008) and 4 fold higher pAkt(Ser473) compared with 231 IR-B OE cells (p=0.0016). Further, we found that 231 IR-A OE cells had approximately 2 fold greater expression of c-myc protein compared with both 231 control (p=0.047) and 231 IR-B OE cells (p=0.026). No differences in c-myc expression were observed between 231 IRB OE and 231 control cells. In our previous studies we found that insulin stimulates c-myc expression and silencing the IR reduces c-myc expression in cancer cells. Our current studies show that IR-A, rather than IR-B is the insulin receptor isoform that regulates c-myc expression in human TNBC. **Reference**: (1) Belfiore et al., Endocr Relat Cancer. 2011; 18(4):R125-R147. (2) Ferguson et al., Breast Cancer Res. 2012; 14(1): R8.

Pediatric Endocrinology
PEDIATRIC ENDOCRINE CASE REPORTS I
SLC29A3 Pathogenic Variant as a Causal for Dysosteosclerosis, a Poor Osteoclast Form of Osteopetrosis

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SAT-080
Background: Dysosteosclerosis (DSS) is a rare autosomal recessive form of osteopetrosis characterized by metaphyseal osteosclerosis and platyspondly. At the histopathological level, a paucity of osteoclasts has been described when the disease presents genetic heterogeneity. SLC29A3 encodes a nucleoside transporter, that colocalizes intracellularly on the lysosomal membrane, is essential for lysosomal function and which is expressed in osteoclasts. Clinical case: We present a female patient from a nonconsanguineous camerunense family with DSS. Her first fracture at the age of 1 year occurred in the proximal right humerus following a very mild trauma. Subsequently she was referred to our clinic at the age of 1.8 months. At the last evaluation in our clinic at the age of 5.6 years she had suffered 5 femur fractures, 2 humerus fractures and a tibia/fibula fracture. XRays shown cortical thickening and widening of the diaphysis of the long bones, cranial base sclerosis, broad ribs with sclerosis and platyspondyly. Bone marrow biopsies and bone biopsy were not performed. The subject did not have any neurological symptoms, dental anomalies or present any dermatological issues. Dual-energy X-ray absorptiometry of the individual showed a lumbar spine BMD Z-score of +8 at 20 months of age, +7.1 at 3 years 5 months, +6.4 at 4 years 6 months and +5.1 at 5 years 6 months of age. TRACP 5 B 4.7 U/L is low in concordance with an osteoclast poor osteopetrosis, and the RANKL>>2.1 ipMol/L is high. On the other hand, CTX 1.298 ng/mL is normal. We identified and confirmed the homozygous pathogenic variant p.Arg386Gln in SLC29A3 using whole exome sequencing and sanger capillary electrophoresis.Conclusion: In summary, we show that a homozygous change in exon 6 of SCL29A3 (NM_00074; c.1157G>A; pArg386Gln) results in a phenotype of Dysosteosclerosis, adding a case to the small group of patients were pathogenic variants in this gene have been described causing sclerosing bone dysplasias with hallmarks of dysosteosclerosis. Interestingly, the other DSS cases and H syndrome cases confirms the clinical heterogeneity of disorders caused by mutations in SLC29A3 and the lack of a clear genotype-phenotype correlation. Phenotypically is also interesting to show that in our patient the bone mineral density has been decreasing over the time, with the concording increasing of TRAB5b. More mutations and functional investigations are needed to get a clearer picture for SCLC29A3 and how is involve in the process of osteoclast differentiation and activity.

Cardiovascular Endocrinology
PATHOPHYSIOLOGY OF CARDIOMETABOLIC DISEASE
Follistatin-Like 3 (FSTL3), a Transforming Growth Factor β (TGFβ) Ligand Inhibitor, Regulates Placental Development in Mice

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SUN-552
Follistatin-like 3 (FSTL3), a glycoprotein that inhibits transforming growth factor-β (TGFβ) ligands such as activin, is expressed highly in the placenta and other vascular tissues. In addition, FSTL3 is strongly induced in pre-eclamptic placenta. To test the hypothesis that FSTL3 function is required for capillary bed structure and function we studied the placenta in FSTL3 gene deleted mice (FSTL3 KO). We have previously shown that FSTL3 deletion produces striking defects in the placenta when compared to WT. Placental size increases significantly in comparison to WT, at 16.5 and 18.5 dpc, with concurrent reduction in placental efficiency at 18.5 dpc. Histological analyses reveal structural differences in placental junctional zones in FSTL3 KO placenta compared to WT. Morphometric analyses show that the labyrinth area compared to the placenta area is significantly reduced in FSTL3 KO mice. We also found that activin-responsive FSTL3-synexpression genes are upregulated in FSTL3 KO placenta. Of these, EPHB4 protein is induced in the placenta along with its ligand EphrinB2. Here we show that FSTL3 deletion leads to endothelial cell expansion but reduction in blood vessel density along with increased extracellular matrix deposition. Further investigation of the placental phenotype revealed differential expression patterns of desmin and cytokeratin protein, reduced von Willebrand factor (VWF) and increased CD31 and VEGFR2 labelling within FSTL3 KO mice placental labyrinths. To identify mechanisms that might lead to the altered placental development in FSTL3 KO mice qPCR analyses were performed. Our results identified differences in the expression of crucial transcripts, such as *Cdh5, Pgf, Fra1* and *Cited1*, that are associated with the regulation of vascular biology. Additionally, we find increased Histone3 and SMAD2 phosphorylation in FSTL3 KO placenta indicating