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عنوان المشروع باللغة العربية - Title of the proposed project in Arabic	دور طفرات جين PLS3 في هشاشة العظام في السعودية
Title of the proposed project in English	Contribution of PLS3 mutations to Osteoporosis in Saudi population.
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التخصص الدقيق للمشرف الرئيس - Specialty of PI	Clinical Biochemistry
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المدة المتوقعة لإنجاز البحث منذ الحصول على موافقة عمادة الدراسات العليا (بالشهور) - Expected time in month to finish	شهر 12
Abstract of the proposal (No more than 200 words)	Plastin 3 (PLS3), a protein involved in the formation of filamentous actin (F-actin) bundles, appears to be important in human bone health, variants in PLS3 have found in the patients with X-linked osteoporosis and osteoporotic fractures. To the best of our knowledge the mutational analysis of PLS3 in Saudi osteoporotic patients have not been studied previously. Therefore, current

study is designed to investigate the pathogenic mutations causing osteoporosis and its prevalence in Saudi osteoporotic patients from Riyadh. Direct DNA sequencing will be performed to identify the pathogenic mutations in PLS3 gene, and its further screening in ethnically matched normal control through SNP genotyping. This study will be first step toward the establishment of nationwide genetic screening of osteoporosis in Saudi Arabia. Hence, identification of pathogenic mutations in osteoporotic patients will help to plan treatment and prevention strategies.

Hypothesis of the proposal

- This study will help to screen out the mutant and genetic carrier individuals that will help to develop diagnosis and genetic counseling strategies to prevent genetic osteoporosis in Saudi population.
- Moreover, through seeking pre implantation genetic diagnosis (PGD), families with known mutations may have the possibility of having children free from those mutations.
- This study will help to establish the basis for the nationwide genetic diagnosis program for osteoporosis.

Specific objectives

- To identify the mutations in PLS3 gene causing osteoporosis in Saudi individuals from Riyadh.
- To investigate the frequency of PLS3 mutations and association with other factors in Saudi osteoporotic patients.

Methodology & Major Techniques to be used

Population

Patients attending hospitals in the Riyadh for the complaint of osteoporosis and bone fracture or other bones associated diseases. Family history and anthropometrics will be obtained. Informed written consents will be obtained before blood sampling.

Blood Sampling:

5 ml venous blood samples from 200 osteoporotic patients will be collected in EDTA tubes. Sampling will be performed at different hospitals of the Riyadh. After collection samples will be stored in -20 °C before DNA extraction.

Biochemical Analysis.

Serum will be isolated and performed for the biomarkers associated with osteoporosis including vitamin D, Calcium, Phosphorus, iron, PTH etc.

DNA extraction:

Genomic DNA will be extracted by using Qiagen Genomic Extraction kits, which simply employ spin columns, for the isolation of DNA. The spin columns contain a silica resin that selectively binds DNA, depending on the salt conditions and

other factors influenced by the extraction method.
Genetic screening:
PLS3 will be amplified by specially designed primers through conventional polymerase chain reaction conventional thermocycler (Applied Biosystems, Foster City, CA). PCR product will be purified by treating with Shrimp alkaline phosphatase enzyme. All PCR products will be subjected to direct DNA sequencing with Big Dye Terminator version 3.1 (Applied Biosystems, Foster City, CA). Sequenced samples will be electrophoresed on an ABI 3730 genetic analyzer, and the traces will be inspected by SeqMan software (DNASTar Lasergene version 5.0.221.0). Pathogenic mutations/polymorphisms of PLS3 in ethnically matched controls will be investigated by SNP Genotyping Analysis Using TaqMan Assays. Special fluorescent probed primers will be designed and amplified by realtime PCR (Applied Biosystems, Foster City, CA).

Availability of Samples

Yes

Availability of Chemicals

Yes

Availability of Instruments

Yes

Ethical Approval

Ethical approval is available

Recent References

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