



Technology Sheet

R.E.M.S. (Radiofrequency Echographic Multi Spectrometry)

Echolight has developed the very first non-invasive solution for bone strength assessment and early diagnosis of Osteoporosis. EchoS is a breakthrough ultrasound device for bone characterization and micro-architecture assessment through an innovative approach that enables the scanning of central reference sites (lumbar vertebrae and proximal femur). EchoS is the only solution capable of combining the advantages of the two main existing technologies (DXA and QUS), allowing our approach to bring axial bone densitometry at the point of care, with a significant beneficial impact on current diagnostic protocols and subsequent patient management. This will also open concrete perspectives for future worldwide standardization of intervention thresholds on the basis of more objective and reliable criteria, increasing the accuracy of Osteoporosis diagnosis.

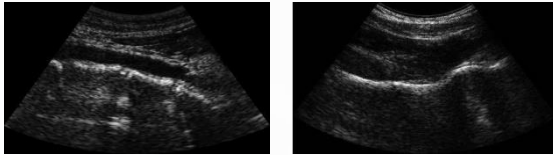
EchoS technology is based on the new and proprietary R.E.M.S. (Radiofrequency Echographic Multi Spectrometry) method: an innovative ultrasound (US) approach to the diagnosis of Osteoporosis, which integrally exploits all the spectral features of the "raw" radiofrequency (RF) signals acquired during an echographic scan of the target anatomical site to determine the status of internal bone architecture. The automatic combined analysis of B-mode images and corresponding RF data provides two novel parameters: the Osteoporosis Score (O.S.), which is directly correlated with BMD, and the Fragility Score (F.S.)*, which quantifies the actual bone strength by assessing structural fragility independently of BMD.

(*Fragility Score: to be released soon)



Diagnostic Output

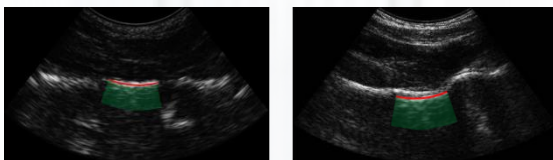
TARGET INTERFACE VISUALIZATION



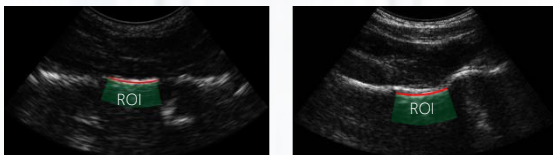
ULTRASOUND ACQUISITION



AUTOMATIC SEGMENTATION OF BONE INTERFACES



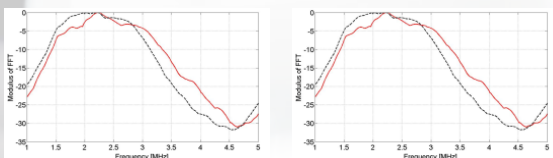
AUTOMATIC DETECTION OF ROIs



AUTOMATIC SIGNALS ANALYSIS



AUTOMATIC SPECTRAL COMPARISON



DIAGNOSTIC OUTPUT

T-SCORE
Z-SCORE
BMD (g/cm^2)
FRAX®
FRAGILITY SCORE*
BODY COMPOSITION INDEX

To perform the diagnostic investigation, the operator should preliminarily visualize the first target interface (i.e. vertebra L1 for lumbar acquisitions or femoral neck for hip scans) and set image depth and focal position to have the target interface in the central part of the image and in correspondence of the focus. Afterwards, the software-assisted US acquisition starts. During the scan, the algorithm automatically detects the bone interfaces (red lines) and calculates the ROIs for data analysis (green areas). The automatic data processing is then started, including RF signal analysis and spectral comparison with reference models for calculation of diagnostic parameters and generation of the final medical report.

R.E.M.S. method provides two new numerical parameters: O.S., which directly correlates with BMD measurements (in g/cm^2) and consequently with DXA diagnostic evaluations (BMD; T-Score; Z-Score), and F.S.*, which provides an independent estimate of bone fragility and fracture risk. Therefore, EchoS medical report contains all the common parameters for Osteoporosis diagnosis through bone density assessment: BMD (g/cm^2), T-Score, Z-Score. In addition, F.S.* evaluates the quality of internal bone micro-architecture. Finally, the 10-year risks of osteoporotic fractures (generic/hip) are calculated through the integrated FRAX® software. Each patient can be examined and diagnosed in less than 2 min, ensuring the compliance of the protocol with time constraints of clinical routine.

(*Fragility Score: to be released soon)



Approach

Ultrasound scans are performed by EchoS echographic device equipped with a convex transducer operating at 3.5 MHz, allowing the simultaneous acquisition of conventional B-mode images and corresponding unprocessed RF signals. The scan lasts about 1 min. R.E.M.S. approach is based on the idea that RF signals, acquired during an echographic scan of a target bone district, can be used to determine the health status of the considered bone through advanced comparisons with previously derived reference spectral models of the possible pathological or normal conditions. This method is natively integrated with US imaging, since, on the

hand, the regions of interest (ROIs) for diagnostic calculations within the investigated bone are automatically identified exploiting both morphologic details and RF spectral features, and, on the other hand, the simultaneous acquisition of several RF scan lines for each image frame provides a solid and reliable statistical basis for subsequent spectral processing. Data analysis includes the calculation of the O.S. value and the F.S. value, based on the correlation between frequency spectra of acquired RF signals and the appropriate reference models of "osteoporotic" and "frail" bones, respectively. BMD, T-score and Z-score are then derived from the O.S. value.

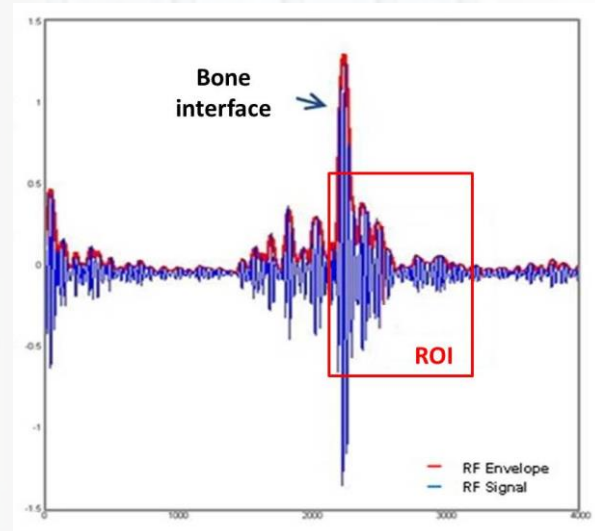
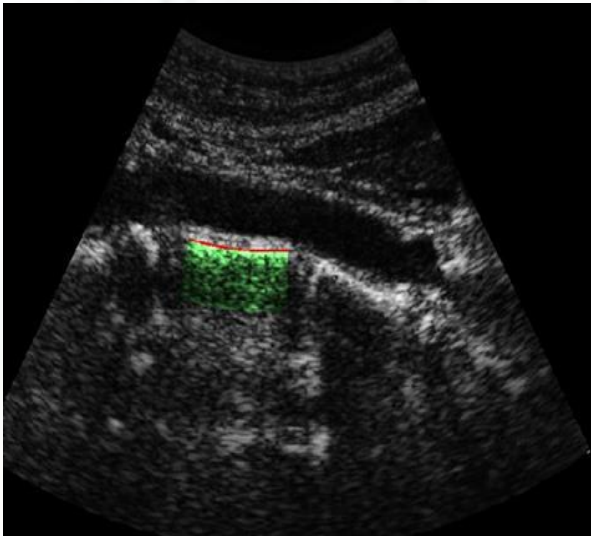
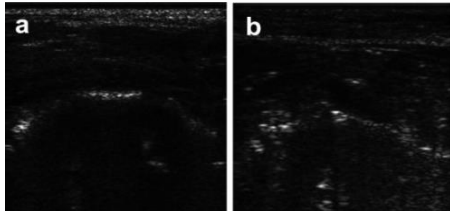
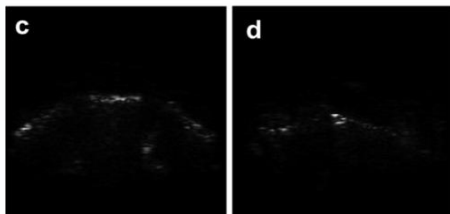


Image Analysis

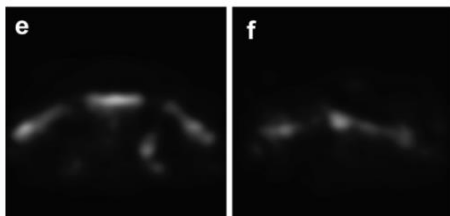
Automatic Identification of Target Bone Structures: the first operation carried out by the algorithm is the automatic identification of the target bone interfaces within the sequence of echographic images acquired on the patient considered. This is achieved by performing the following steps on each acquired frame:



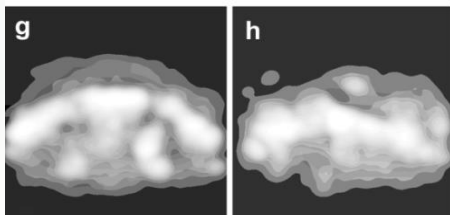
(a,b) Rearrangement of image data in rectangular matrices from the original sectorial images: (a) sample echographic image containing a vertebral interface that will be automatically identified, (b) sample "noisy" echographic image that will be automatically discarded because there are no suitable vertebral interfaces.



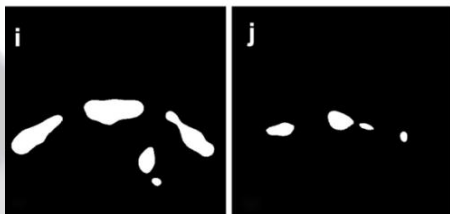
(c,d) Brightness masking.



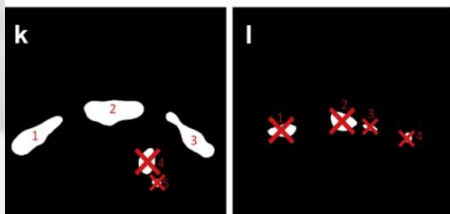
(e,f) Contrast enhancement and image smoothing.



(g,h) Histogram equalization.



(i,j) Thresholding.



(k,l) Morphologic evaluations: (k) clusters 4 and 5 are excluded because they are outside the expected size range for a vertebral interface, whereas clusters 1–3 are retained, and, after finer morphologic evaluations, cluster 2 is labeled as a "possible vertebral interface", whose actual nature will be verified through a dedicated spectral analysis; (l) all the clusters are excluded because they are outside the expected size range, and the frame is discarded.

Spectral Comparison

Diagnostic parameter calculations are actually performed on **RF signal segments** corresponding to specific ROIs, which are automatically selected starting from the coordinates of the identified bone interfaces. The overall concept is that spectra of "raw" RF signals backscattered from human bones during an "in vivo" echographic scan contain useful information about the bone status, including both **quantity** (e.g. BMD) and **quality** (e.g. elasticity) parameters. Accordingly, the O.S. value measures the degree of similarity between RF spectra obtained from the considered patient and spectral models previously derived from subjects with a low BMD (T-score ≤ -2.5), with respect to those derived from normal subjects (T-score ≥ -1.0); on the other hand, the F.S.* value quantifies an analogous spectral similarity to subjects that reported a recent fragility fracture with respect to control subjects without fracture history. In this way, each spectrum belonging to the identified ROIs undergoes an overall shape comparison with two pairs of sex-, race- and age-matched reference models that had been calculated from a database of real subjects.

Osteoporosis Score and Fragility Score

Once all the frames belonging to the US dataset of the patient have been analyzed, the system verifies if the number of detected interfaces is sufficient to obtain a reliable diagnosis and **diagnostic calculations are performed on each RF spectrum of the identified ROIs**. If this minimal frame condition is not satisfied (it happens rarely), the dataset is labeled as "noisy" and the operator is asked to repeat the US scan. For the O.S. calculation, the considered RF spectrum is classified as "osteoporotic" if the value of its Pearson correlation coefficient with the appropriate osteoporotic model spectrum is higher than the corresponding value with the related healthy model spectrum, otherwise, it is classified as "healthy." The percentage of analyzed spectra classified as "osteoporotic" represents the O.S. value of the analyzed ROI. The O.S. values of all the ROIs belonging to the same bone target (i.e. the same vertebra or the same femoral region) are then averaged to obtain the O.S. value of the considered bone structure. **The same evaluations are repeated for each identified bone target, and the final O.S. is the average of the single values.** An analogous procedure is simultaneously applied to calculate the F.S.* value, with the only difference being that "osteoporotic" and "healthy" reference models are replaced by the corresponding spectral models of "frail" and "non-frail" bone structures. An important aspect to be underlined is the **extreme ease of use of the system**: inexperienced operators, who had previously received only a 3-h specific training session, performed acquisitions of suitable quality for diagnostic calculations in 96.3% of cases (i.e. the incidence of "noisy" datasets was limited to 3.7%, and all of them became suitable at a second scan).

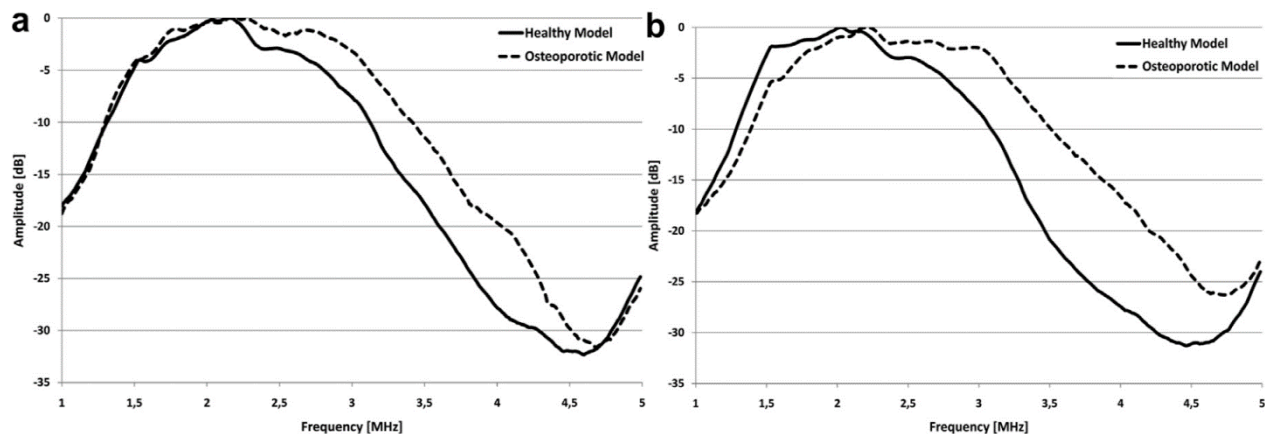
(*Fragility Score: to be released soon)



Statistical Analysis

Selection of the appropriate pairs of spectral models from the reference database takes into account the following patient characteristics: sex, race, age and body mass index (BMI). Current database version includes around 10.000 subjects, uniformly distributed in the age range from 30 to 90 years and covering the whole interval of typical BMI values, from under-weight to obese individuals. Database sample size as a function of considered age ranges was established according to widely adopted literature-reported rules (Osteoporos Int 2008;19:71; Osteoporos Int 2006;17:1283). Subjects were grouped into 5-y intervals and, in each age interval, they were further split into three subgroups based on their BMI (i.e. under-weight/normal-weight, over-weight, obese). For each obtained subgroup, the first 100 individuals were included in the reference database. The subjects were recruited through a multicenter Italian clinical study involving hospitals and clinics specialized in osteoporosis diagnosis and all of them underwent the following diagnostic examinations:

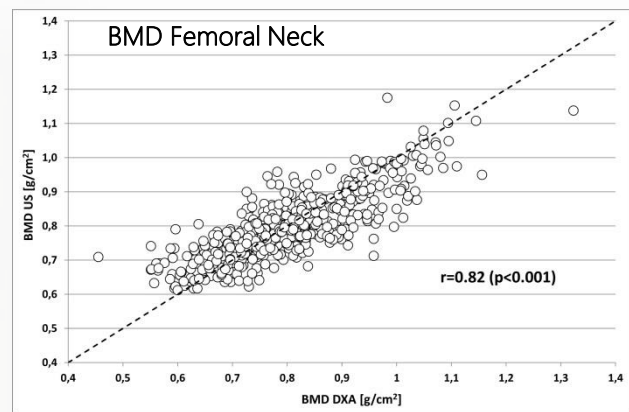
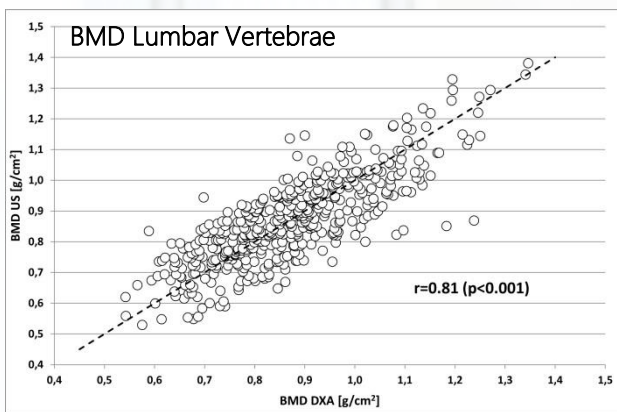
DXA scan of lumbar spine and/or proximal femur, TBS calculation where applicable, echographic scan of the same anatomical districts performed with EchoS system, FRAX[®] questionnaire for the estimation of fracture risk. Therefore, for each subject, the database includes: anonymous personal data (age, sex, race, BMI), DXA and TBS investigation reports, US datasets, and FRAX[®] questionnaire answers. In each identified subgroup, these data were used to calculate the corresponding pairs reference spectral models for the following bone conditions: "osteoporotic/healthy", which is assessed through the O.S. value, and "frail/non-frail", which is assessed through the F.S. value. The dedicated procedure for the derivation of reference spectral models has been detailed in a very recent paper referred to the case of O.S. calculation (Ultrasound Med. Biol. 2015;41:281). All the obtained models were also included in the database and associated to the relevant subgroups.



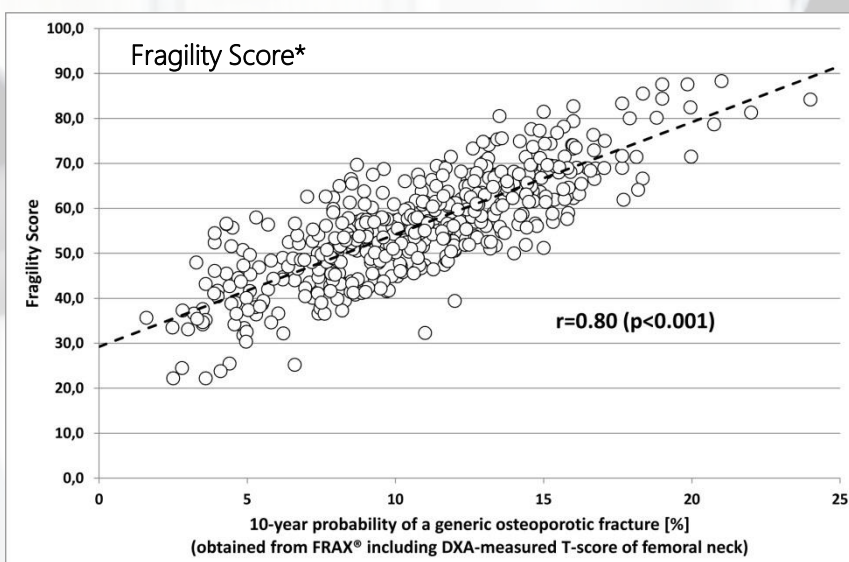
Sample reference model spectra for the calculation of O.S. value on lumbar spine. Pairs of "osteoporotic" and "healthy" spectral models obtained for two different subgroups of female subjects: (a) under-weight and normal weight women aged 51-55 y; (b) under-weight and normal-weight women aged 56-60 y.

Clinical Validation

Diagnostic accuracy of the technology has been clinically tested according to international scientific frameworks *(Osteoporos. Int. 2006;17:1449; Q. J. Med. 2008;101:605). Patients were recruited on the basis of the following enrollment criteria: aged 30-90 y, medical prescription for a spinal and/or femoral DXA, informed consent. Each patient underwent the prescribed DXA scans, EchoS echographic scans of the same anatomical districts, and FRAX® questionnaire for the estimation of fracture risk. BMD values derived from O.S. calculation were compared with the results of DXA measurements, while F.S. values were evaluated against FRAX® predictions of 10-year probabilities of a generic osteoporotic fracture (calculated with the inclusion of DXA-measured femoral neck T-score).



QUANTITATIVE ASSESSMENT OF ECHOS	VERTEBRAE	FEMORAL NECK
SMALLEST DETECTABLE DIFFERENCE (SDD) [g/cm ²]	0.010	0.005
INTRA-OPERATOR REPEATABILITY (RMS-CV) [%]	0.35%	0.25%
INTER-OPERATOR REPEATABILITY (RMS-CV) [%]	0.54%	0.41%
DIAGNOSTIC AGREEMENT WITH DXA	93.1%	94.2%



Agreement between Fragility Score* and FRAX® to evaluate the risk of osteoporotic fractures.

*Fragility Score**, derived from the same US data acquired for bone density assessment, showed a strong correlation with FRAX® fracture risk, whose calculation requires the answers to the FRAX® questionnaire and a DXA scan of femoral neck.

(*Fragility Score: to be released soon)

Conclusions

The peculiar feature of R.E.M.S method is the exploitation of RF signals acquired during an echographic scan of the target bone structure to determine the internal bone architecture through detailed comparisons with reference spectral models. Another important feature of the technology is its full automation, which reduces to a minimum the dependence on operator experience. In fact, the implemented algorithm automatically identifies and discards "noisy" acquisitions, ensuring that diagnostic evaluations are performed only on US datasets reaching a specifically determined quality threshold. The extreme ease of use of the described system has also been demonstrated, together with its compliance with time constraints of

clinical routine, since each patient can be examined and diagnosed in less than 2 min. Because of its accuracy levels, combined with the complete absence of ionizing radiation and the proven ease of use, this method can be effectively employed for diagnosis of osteoporotic disease at an earlier stage through population mass screenings. Moreover, the US assessment of internal bone structure not only gives information related to BMD, but also provides further insights into the structural quality of bone and its real strength, offering a simultaneous, independent and accurate prediction of fracture risk. The integration of all these features makes R.E.M.S method the future state-of-the-art approach for the early diagnosis of Osteoporosis.



Reference Sites (Europe)

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Prof. Jean-Yves Reginster

President ESCEO (European Society for Clinical and Economic Aspects of Osteoporosis and Osteoarthritis). University of Liège and Bone and Cartilage Metabolism Unit Liège, Belgium.

Prof. Cyrus Cooper

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Prof. René Rizzoli

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Gerolamo Bianchi, Giuseppe Girasole.

Azienda Sanitaria Genovese, Dipartimento Apparato Locomotore UO Reumatologia - Ospedale La Colletta, Arenzano (GE)



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Company

Echolight is a high-tech research based biomed company, incorporated in Italy, for the development of innovative technologies in the medical device arena. Our mission is to provide the world medical community with the very first radiation-free and office-based solution for the Early Diagnosis of Osteoporosis. Echolight will make the early detection of Osteoporosis more accurate and easily accessible to meet both clinician's and patient's needs everywhere. Echolight is in compliance with the standard: UNI CEI EN ISO 13485:2012; ISO 13485:2003; CE Mark Medical Device Class IIa.



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