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Research Article

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Time Series Prediction of Plasma Hormone Concentration

Evidence for Differences in Predictability of Parathyroid Hormone Secretion Between Osteoporotic Patients and Normal Controls

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Abstract

Recent evidence links osteoporosis, a disease of bone remodeling, to changes in the dynamics of parathyroid hormone secretion. We use nonlinear and linear time series prediction to characterize the secretory dynamics of parathyroid hormone in both healthy human subjects and patients with osteoporosis. Osteoporotic patients appear to lack the periods of high predictability found in normal humans. Our results may provide an explanation for why an intermittent administration of parathyroid hormone is effective in restoring bone mass in osteoporotic patients. (*J. Clin. Invest.* 1995. 95:2910–2919.) Key words: parathyroid hormones • osteoporosis • bone remodeling • osteoclast • osteoblast

Introduction

Fluctuations in parathyroid hormone (PTH)[1] plasma concentration over short time intervals seem to play an essential role in maintaining the physiological balance of bone resorption and bone formation (1–4). The concentration of PTH in the bloodstream of healthy human subjects fluctuates in an episodic, pulsatile manner with a mean PTH pulse frequency between 1 pulse per hour (large pulses) and 1 pulse per 10 min (small pulses) (1). In the physiological bone remodeling process, bone resorption by osteoclastic cells and bone formation by osteoblastic cells are functionally coupled (5). PTH has been shown to act directly on osteoblastic cells whereas its action on osteoclastic cells is mediated by local factors (6–10). Animal experiments have demonstrated that an intermittent administration with daily injections of PTH increases bone mass and normal connectivity, whereas a continuous administration by infusion at the same mean rate leads to a net loss of bone mass and of bone structure (11–13). The bone becomes more porous, as seen in osteoporotic patients, suggesting that this disease is caused by a disruption of the normal temporal dynamics of PTH secretion. In accordance with these results, a therapy protocol with daily injections of PTH produced the largest gain in trabecular bone mass in osteoporotic patients (14, 15).

The evidence linking the secretion of PTH to bone remodeling suggests that secretory patterns in osteoporotic patients and healthy subjects may be different. Until recently, a comparison of the dynamics of PTH secretion in healthy and osteoporotic subjects was not possible because data at a sufficiently fine temporal scale were not available. In a recent experiment (1), we measured PTH serum concentration at 2-min intervals in both healthy and osteoporotic subjects (Fig. 1). These data now offer a unique window into the dynamics of PTH secretion in both subject groups.

Traditional approaches to identifying dynamical disease states have involved examination of the time series and power spectra of physiological variables for evidence of changes in periodicity or regularity of a process (16, 17). Unfortunately, healthy subjects cannot be distinguished from patients with osteoporosis using the mean or variance of PTH serum concentration (Fig. 1) or the power spectrae (Fig. 2). These classical techniques for time series analysis do not appear well suited to discovering differences in the dynamics of such irregular time series.

Time series prediction has proved effective in characterizing irregular complex time series and separating deterministic (chaotic) behavior from some forms of random behavior (18–20). Most of this work has focused on distinguishing chaotic behavior from zero order (independent identical distributed) and first order (the series of first differences is independent identical distributed) stochastic processes, and more complex forms of correlated processes have not been extensively analyzed. Recent work shows that such a predictive model is particularly effective when applied to short time series that contain on the order of a few hundred data points (19, 20). Systematic differences in the dynamics may lead to very different degrees of predictability. Such a predictive model has recently been applied to time series analysis of electroencephalogram (EEG) data (21).

We describe a technique for identifying differences in the dynamics of PTH secretion between healthy subjects and patients with osteoporosis using time series prediction. A single predictor trained on pooled data from several healthy subjects as well as predictors individually trained on each time series were used to predict time series from both healthy and osteoporotic subjects. Differences in the rate at which predictions diverged from the true evolution of the time series were used to divide healthy subjects into groups exhibiting states of low

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1. *Abbreviations used in this paper:* AR, autoregressive; ARIMA, autoregressive integrated moving average; EEG, electroencephalogram; I, integrated; MA, moving average; PTH, parathyroid hormone.

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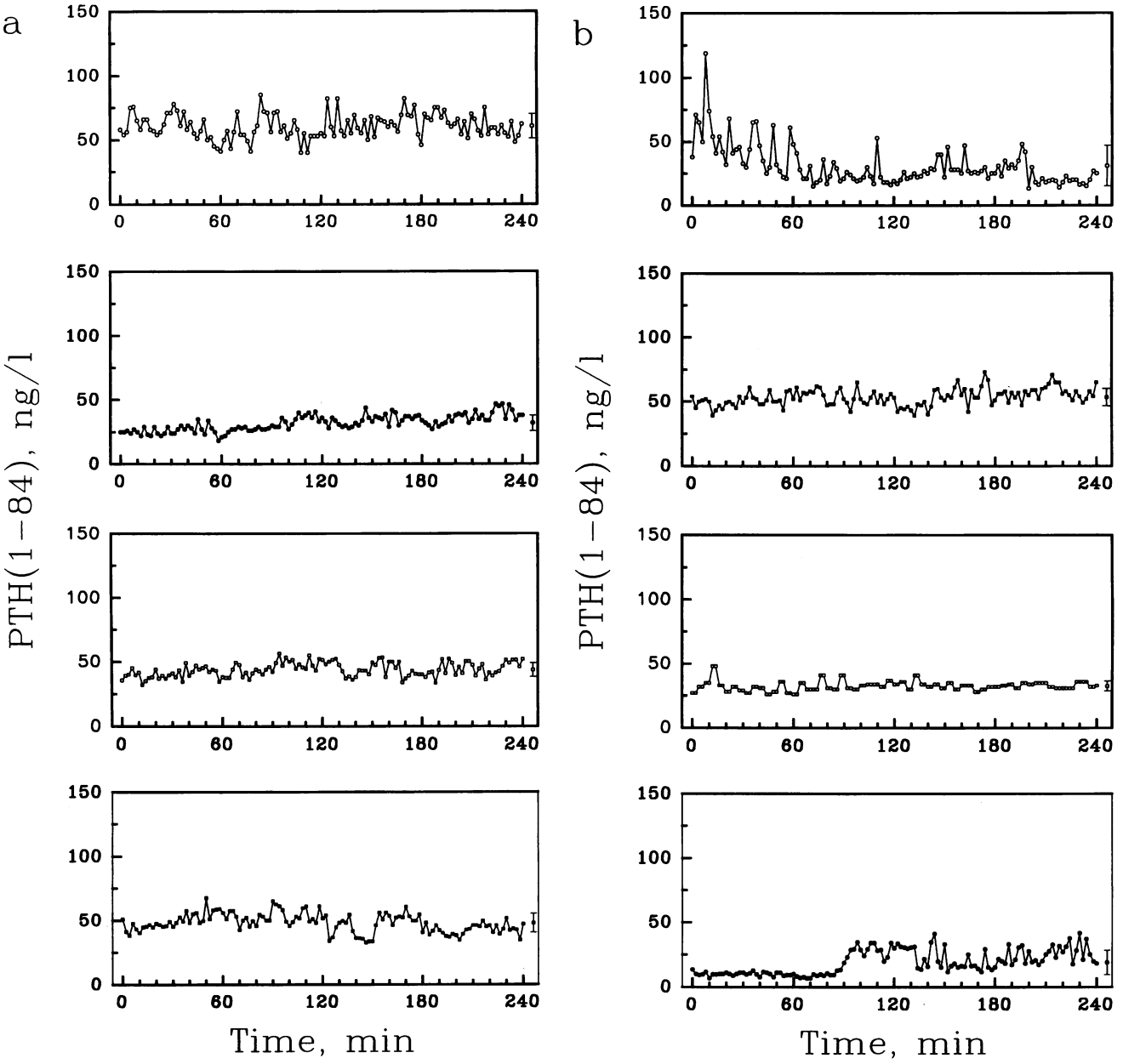

Figure 1. Representative time series of PTH(1-84) serum concentration (4-h period). (*a*) Four healthy subjects (total of nine subjects), mean PTH serum concentrations: (○) 60.5±9.5 ng/liter; (■) 47.8±7.4 ng/liter; (□) 43.7±5.5 ng/liter; (●) 31.9±6.1 ng/liter; (*b*) Four osteoporotic patients (total of six patients), mean PTH serum concentrations: (■) 53.3±6.7 ng/liter; (□) 32.6±3.9 ng/liter; (○) 30.9±15.9 ng/liter; (●) 18.8±9.4 ng/liter. Whether a PTH serum concentration time series is from a healthy subject or an osteoporotic patient cannot be determined from the mean concentration nor from the standard deviation.

and high predictability. Examination of longer time series from several healthy subjects suggests a tendency to switch between these states of low and high predictability. Osteoporotic patients exhibited divergence rates similar to the healthy subjects in the low predictability group, and none showed any evidence of the highly predictable behavior found in some healthy subjects. These differences could not be found using "classical" methods of time series analysis. Our findings suggest, that osteoporosis may be a disease in which the dynamics of hormonal fluctuations is altered in a subtle but critical manner.

Methods

Subjects. Twelve healthy men (aged 24–42 yr), three women with postmenopausal osteoporosis (aged 55–62 yr), and three men with idiopathic osteoporosis (aged 31–42 yr) took part in this study. The studies reported were approved by the local Committee on Medical Ethics, and all subjects gave their informed written consent. All healthy subjects had an unremarkable personal and family medical history. Physical examination, electrocardiogram, white blood count, differential, protein, creatinine clearance, albumin, total calcium, sodium, potassium, magnesium, chloride, creatinine, triglycerides, cholesterol, glucose, and nitrogen were normal in both groups. In addition to parameters to exclude secondary osteoporosis and other diseases, we measured alkaline phosphatase, osteocalcin, calcium, ionized calcium, and phosphate. Body weight, diet, and daily physical activity were comparable between groups. In the patients with osteoporosis bone density was assessed by single-energy computed tomography (the mean value was 49% of reference) and dual-energy computed tomography (the mean value was 52.6% of reference). Lateral spine (thoracic and lumbar) x-rays were

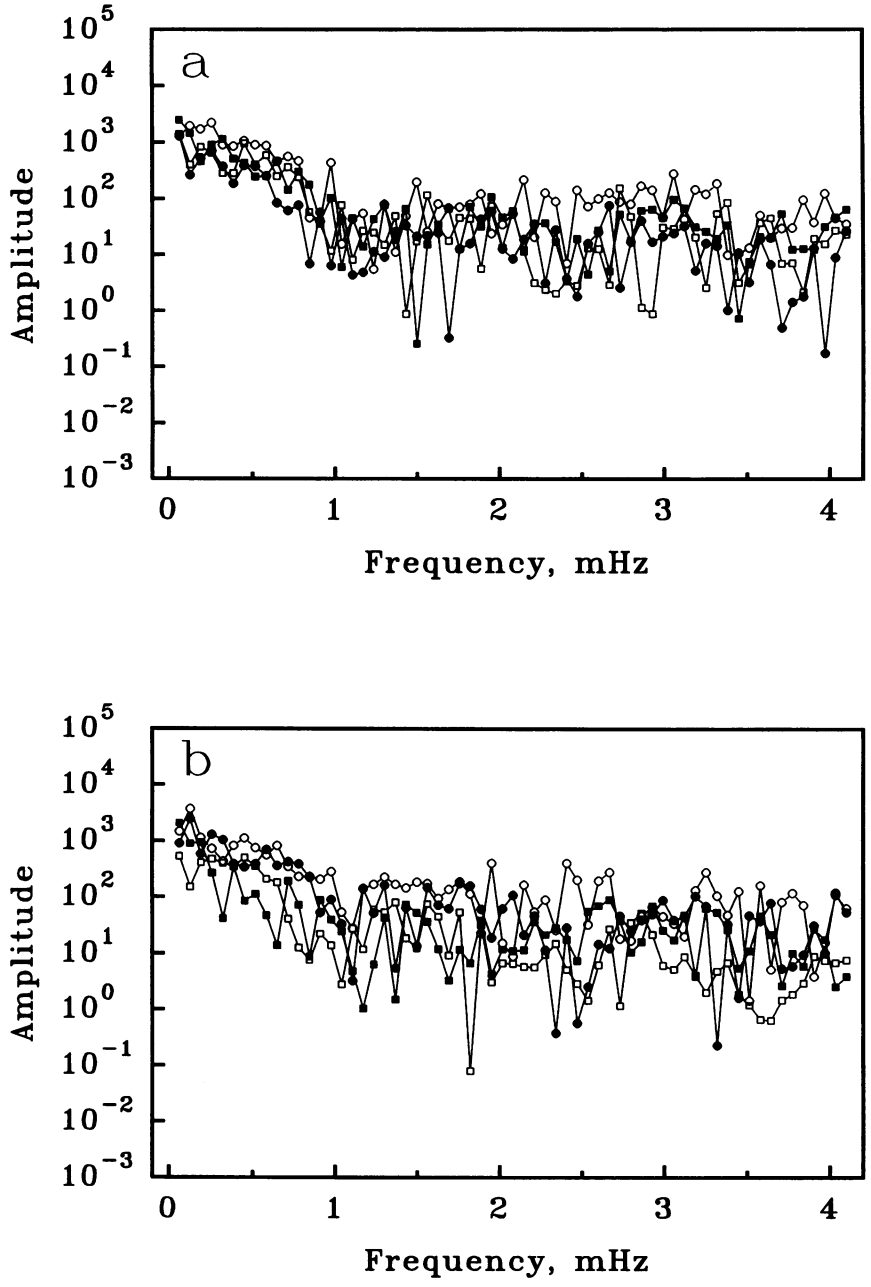

Figure 2. Power spectra of the time series from Fig. 1. (*a*) Healthy subjects; (*b*) osteoporotic patients. All spectra of both groups are broad and quite flat up to the Nyquist frequency (4.17 mHz), characteristics common to stochastic and chaotic dynamic processes. None of the spectra exhibits strong spectral features that would indicate strong periodic behavior and could be easily used for reliable classification.

obtained from all patients. The typical configuration of osteoporotic vertebral bodies was present in all subjects. Vertebral fractures, i.e., height reduction of > 25% was present only in two of the patients. Some of the subjects in this study as well as the details on design and measurements are described in more detail in a previous publication (1).

Sampling protocol. In three of the healthy subjects, PTH serum concentrations were measured in blood samples taken every 2 min over an extended period (21 h for one subject and 24 h for the other two). In the remaining nine healthy subjects and the six osteoporotic patients, PTH serum concentrations were also measured at 2-min intervals, but over much shorter periods of time (3.5–9 h). All PTH concentrations were measured in duplicate using either a two-site chemiluminometric (sandwich) immunoassay or an intact PTH immunoradiometric assay. Blood samples (1 ml) were drawn via a central venous catheter. The specimens were then centrifuged at 4°C and frozen at −20°C within 45 min and stored for 1–3 mo. 24 h before sampling no alcohol intake, medication, or caffeine was permitted. The central venous catheter was placed 1 h before the initiation of blood sampling. Throughout the study, the subjects rested in bed.

Measurement of PTH serum concentrations. The PTH concentrations of all serum samples were measured in duplicate. To avoid interassay variations, all samples from an individual subject were analyzed in the same assay. The 21- and 24-h PTH serum concentration time series and half of the other PTH time series were measured using a two-site chemiluminometric (sandwich) immunoassay (Magic Lite Intact PTH; Ciba-Corning Diagnostics Corporation, Medfield, MA; intra-assay coefficient of variation [CV] 3.4%, interassay CV 4.3%). The other half of the results were obtained with the Allegro intact PTH immunoradiometric assay system (Nicols, San Juan Capistrano, CA; intra-assay CV 5.1%; interassay CV 7.8%). Exactly one third of the samples of normal subjects and one third of the samples of osteoporotic patients were analyzed with the chemiluminometric immunoassay and in both groups two thirds with the immunoradiometric assay.

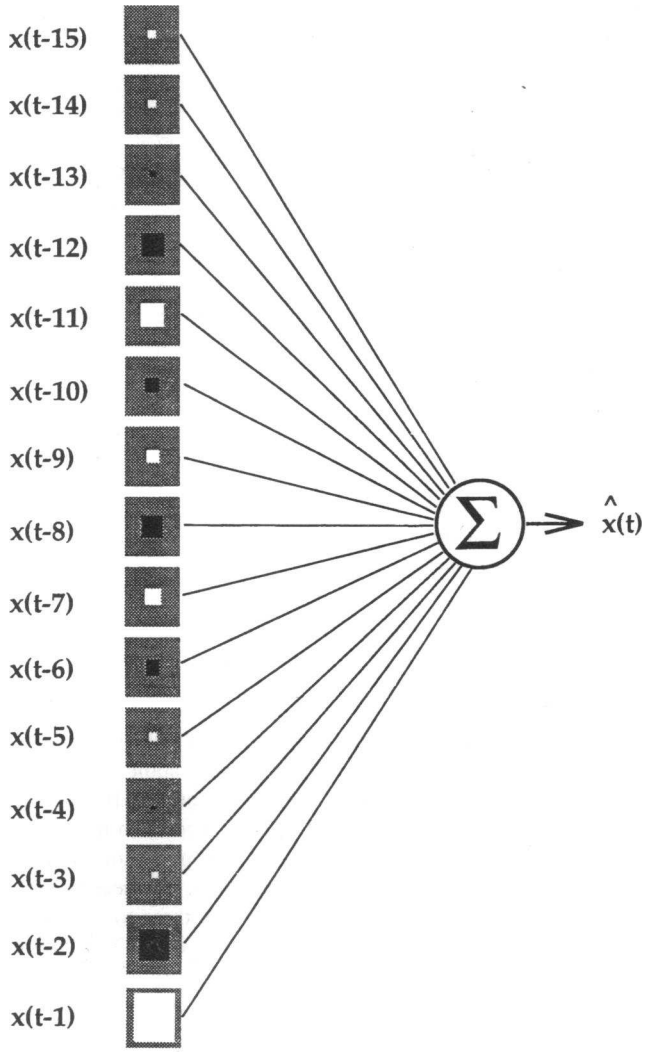

Figure 3. Architecture of the prediction network. The estimate of the current value of the time series ($\hat{x}(t)$) is the weighted sum of the previous 15 values of the time series. Gray squares represent the weights of the network related to the samples of the time series ($x(t-15), \ldots, x(t-1)$). The size of the white or black square inside each gray square indicates the magnitude of the weight assigned to that sample (black squares: negative weights, white squares: positive weights).

Time series prediction. In this approach to time series analysis, we find models that attempt to predict the next value $x(t_i)$ of a time series from a number of previous values:

$$x(t_i) = f(x(t_{i-1}), x(t_{i-2}), \ldots, x(t_{i-m})) + \epsilon_i,$$

where $x(t_{i-1})$ is the value at time t_{i-1}, m is the number of previous values used for prediction, and ϵ_i represents noise or fitting error. The model f is selected by minimizing some measure of misfit (such as mean square error) over a set of training examples drawn from observed samples of the time series. When the predictive model is linear ($f(x(t_{i-1}), \ldots, x(t_{i-m})) = \Sigma\alpha_k x(t_{i-k})$), this type of modelling is equivalent to fitting an autoregressive (AR) model of order m to the data. By modeling the series of first differences using a similar form of predictor we can also fit autoregressive integrated moving average (ARIMA) models, and nonlinear extensions of these models, to a data set.

In this study we used feedforward networks to predict future values of the time series of PTH serum concentration (Fig. 3, Table I). This

Table I. Coefficients of the Predictive Model

Coefficient	Weight value
$x(t-1)$	1.339
$x(t-2)$	−0.350
$x(t-3)$	0.022
$x(t-4)$	−0.004
$x(t-5)$	0.028
$x(t-6)$	−0.102
$x(t-7)$	0.139
$x(t-8)$	−0.177
$x(t-9)$	0.065
$x(t-10)$	−0.073
$x(t-11)$	0.266
$x(t-12)$	−0.217
$x(t-13)$	−0.013
$x(t-14)$	0.029
$x(t-15)$	0.046
Bias	0.566

Coefficients with values less than 0.1 are not significant and may be removed from the model without qualitatively effecting the results of predictions on the test subjects.

form of a predictive model was chosen because it is relatively easy to control over-fitting using regularization functions and cross-validation and such models have proven to be effective predictors for other short, noisy time series (22–24). This class of model implicitly includes the classes of moving average (MA) and AR models (25).

Networks were trained to predict a single time step into the future. To predict multiple steps into the future the single-step map was iterated by feeding back the output of the network into its input. If the predicted value one step into the future is

$$\hat{x}(t_i) = f(x(t_{i-1}), x(t_{i-2}), \ldots, x(t_{i-m})),$$

then the predicted value two steps into the future is computed as

$$\hat{x}(t_{i+1}) = f(\hat{x}(t_i), x(t_{i-1}), \ldots, x(t_{i-m+1})).$$

This process can be repeated to predict any number of steps into the future. The correlation between observed (x_i) and predicted ($\hat{x}_i$) values was measured using

$$arv = \frac{\langle (x_i - \hat{x}_i)^2 \rangle}{\sigma^2(x_i)},$$

where *arv* is the average relative variance, the angle brackets denote an average over all values of the indexed variable, and $\sigma^2(x_i)$ denotes the variance of the indexed variable. This statistic is one measure of the regularity of a time series, and other forms of regularity measure have previously been used to discriminate among hormone secretion time series (26).

To improve the signal-to-noise ratio the raw PTH time series were filtered before prediction by an acausal filter (27) (rolloff frequency $\eta = 0.002$), which does not affect certain nonlinear measures. To avoid any bias due to the filtering process, we selected this particular type of filter because we also used neural network predictors with nonlinear activation functions.

Training and testing of predictive models. Before selecting a distinct network architecture for our detailed analysis, we explored a large variety of different network architectures, applying linear as well as nonlinear sigmoidal feedforward networks with 1–30 input units, 0–50 hidden units, and 1 output unit. A regularization technique (23) was used to control overfitting to the training data, with the weight assigned to the regularization term chosen by cross validation. The value for the regularization term varied from 10^{-5} for the nonlinear networks to 10^{-10}

for the linear networks. The weights were updated by a conjugate gradient descent method. (For a linear model, this fitting technique is formally equivalent to a least-squares linear fit.)

Networks were trained using pooled time series data from the three healthy male subjects whose PTH serum concentrations were measured every 2 min for an extended period. Furthermore we trained our networks using pooled time series data from the osteoporotic patients. Using such a network for testing the osteoporotic subjects we used a "leave one out technique," where the time series actually being tested is left out of the training procedure. The performances of these networks were compared on a validation set that was not used during training and the architecture with the best performance was a feedforward neural network with one layer of weights, linear activation functions, 15 input units, and 1 output unit (Fig. 3, Table I). This network is equivalent to a 15th order AR model, however the regularization procedure yields a final model with only seven significant coefficients (Table I). This model was also the best predictor of the PTH time series in the 15 test subjects and also the best discriminator between the normal and osteoporotic subjects of the 76 different models of various orders that we fit to the data. Furthermore we used the 15 input units–1 output unit network as well a variety of other linear and nonlinear network architectures to perform individual training and testing of each normal and osteoporotic time series. To account for possible nonstationarity in our data (Fig. 1) we also fitted the best predictive ARIMA model individually to the first two thirds of each time series using a variety of different AR, I, and MA orders, which were identified with the help of the autocorrelation and partial autocorrelation function (using the MATLAB System Identification Toolbox; The Mathworks, Inc., Natick, MA). After fitting, the last third of each time series was predicted by the fitted ARIMA model. Furthermore we fitted the best predictive ARIMA model to the pooled PTH data from the three healthy subjects and evaluted the predictive ability on each of the nine remaining time series from normal subjects and six time series from osteoporotic patients. We then compared the predictive results to those obtained from individual training and testing of each time series and also to training on pooled data using the neural network approach.

The 15 input units–1 output unit network was used for most of the simulations discussed in the remainder of this paper. It was used to predict the time series from an additional 15 test subjects; 9 healthy subjects and 6 patients with osteoporosis.

Our use of a single predictive model for all time series differs from other studies, which used a different predictor for each time series (19, 22, 24). We chose this approach because we were primarily interested in discriminating the PTH secretory patterns of healthy subjects from patients with osteoporosis on the basis of relative predictability rather than estimating the absolute degree of predictability in normal PTH concentration time series. However, we performed additional experiments in which each time series was trained (on the first two thirds of each time series) and tested (on the last third) with a single individual neural network predictor using linear as well as nonlinear activation functions.

The arv was computed for each of the 15 test subjects as a function of the number of steps predicted into the future (Table II). The number of prediction steps after which the arv exceeded 0.5 was recorded for each time series. This number is a measure of the number of prediction steps into the future for which the network is useful as a predictor for the system dynamics. An arv of 1.0 may be achieved simply by always guessing the mean of the time series as the predicted value, so a predictor that meets our criterion is on average twice as good as predictor that entirely ignores the dynamics of the time series.

To investigate the possible sensitivity of our prediction method to the frequency at which the PTH serum concentration was measured, we repeated all our prediction simulations using every 2nd and 5th data point of the original PTH concentration time series (4- and 10-min intervals, respectively).

Results

Evidence for states of low and high predictability. Using the 15 input unit–1 output unit neural network trained on pooled data from healthy subjects, we found that the PTH serum concentration time series of the group of healthy subjects exhibits two different types of behavior. In four subjects, the time series could be predicted between 8 and 12 time steps into the future before the arv exceeded 0.5 (Fig. 4). This group of subjects exhibited high predictability. In the remaining five healthy subjects, the time series could be predicted only three to five steps into the future before an arv of 0.5 was exceeded. This group of subjects exhibited low predictability. To verify the existence of these two groups, we clustered the data using a *k-means* procedure and compared the fit of the resulting two normal component model to a single normal fit, using a likelihood ratio test. The two component model provided a significantly better fit than a single component model ($P < 0.03$).

Table II. Number of Prediction Steps (2 min) Until the Prediction Error (arv) Reaches a Value of 0.5

Subject	Prediction step
Normal1	12
Normal2	11
Normal3	8
Normal4	11
Normal5	4
Normal6	5
Normal7	5
Normal8	4
Normal9	3
Osteoporotic1	2
Osteoporotic2	4
Osteoporotic3	4
Osteoporotic4	2
Osteoporotic5	3
Osteoporotic6	6

The linear 15 input–1 output neural network was trained on pooled data from healthy subjects.

We used the two-component model to classify the predictability of the osteoporotic time series and found that all of the osteoporotic patients exhibited low predictability under this classification criterion. The PTH serum concentrations from the osteoporotic subjects could be predicted only two to six time steps into the future before the arv exceeded 0.5. Significant correlations between the age of the subjects, their mean PTH serum concentration, and the predictability could not be found. These categories of predictability could not be identified by computing the mean or the variance of the PTH serum concentration time series (Fig. 1) nor by computing the power spectrum of each time series (Fig. 2).

We tested the significance of differences in predictability between various groups of test subjects using a Mann-Whitney-Wilcoxon rank test (Table III). There was evidence that the nine normal subjects as a group were more predictable than the osteoporotic patients. However, there was much stronger evidence that the four normal subjects falling in the highly predictable category were more predictable than either the osteoporotic patients or the normal subjects falling in the low predictability category. This suggests that although high predictability may be an indicator of health, low predictability alone is not necessarily a strong indicator of disturbances in PTH secretion.

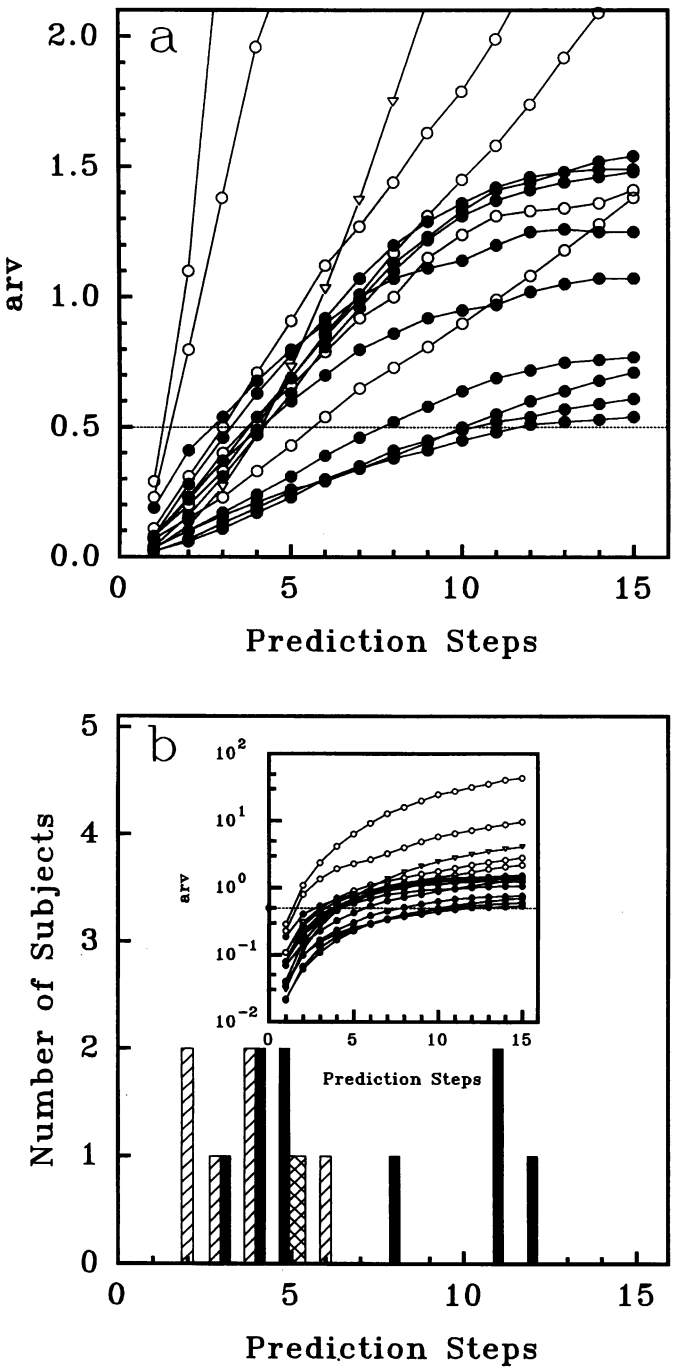

Figure 4. Summary of predictability for healthy subjects and osteoporotic patients. (*a*) arv (prediction error) vs. the number of prediction steps (2-min interval): healthy subjects (●), osteoporotic patients (○), colored noise (▽) with a power spectrum matched to the training data. The dashed line indicates an arv of 0.5 used as the criterion for separating the healthy subjects from the osteoporotic patients. For clarity, only values with an arv < 2.0 are shown. The entire data set is shown in the *inset* in *b*. (*b*) Histogram of the number of subjects vs. the prediction step (2-min interval) at which the arv exceeds a value of 0.5. The *inset* shows the arv (on a logarithmic scale) vs. the number of prediction steps for all subjects (see detail in *a*).

There were also qualitative differences in the graphs of arv versus the number of prediction steps between the healthy subjects and osteoporotic patients (Fig. 4 *a*). All of the healthy subjects showed a "saturation" of the arv as the number of prediction steps increased, although this saturation occurred above an arv of 1.0 for subjects exhibiting low predictability. The osteoporotic subjects and the time series of colored noise showed, with one exception, a linear increase of the arv with the number of prediction steps.

Further analysis of the long PTH concentration time series from the healthy subjects showed that regions of low and high predictability could be found within each of the long time series (Fig. 5). The arv value was computed for a continuously running 60 data points (2 h) window for the entire PTH time series and recorded for all of the longer PTH time series for the 1–15 time steps ahead prediction (Fig. 5 *a*), as well as the time step where the prediction error arv reached a value of 0.5 (Fig. 5 *b*). The time series were then divided into segments at points where the predictability (in prediction steps) was less than 1 SD from the mean predictability across the entire time series (low predictability). This procedure divided each time series into segments of high and low predictability.

The periods of high predictability were on average more common, although the exact proportion of regions of high and low predictability varied among the long time series (10% low, 21% low, and 23% low). The length of the periods of low predictability also showed considerable variation (2 h 24 min to 4 h 55 min). The short time series from the nine healthy

Table III. Differences in Predictability for Various Groups of Subjects

Group 1	Group 2	Significance
Normals	Osteoporotics	$P < 0.03$
(n = 9)	(n = 6)	
7.0±3.5	3.5±1.5	
Pred. normals	Nonpred. normals	$P < 0.01$
(n = 4)	(n = 5)	
10.5±1.7	4.2±0.8	
Pred. normals	Osteoporotics	$P < 0.01$
(n = 4)	(n = 6)	
10.5±1.7	3.5±1.5	

The mean±SD of the number of steps required to reqch an arv of 0.5 is shown for each group. The final column shows the significance of the hypothesis that the number of prediction steps required to reach an arv of 0.5 is greater in group 1 than in group 2. All tests are based on a nonparametric Mann-Whitney-Wilcoxon test (U statistic).

subjects and six osteoporotic patients did not contain enough data points to reliably identify distinct regions of low and high predictability within these time series.

Sensitivity of results. In the following, Mann-Whitney-Wilcoxon rank tests were used to obtain the statistical results. Using the 15 input units–1 output unit neural network as an individual predictive approach, the degree of predictability in the healthy group was significantly ($P < 0.05$) lower (3.9±1.2 time steps) than in the case of the predictor trained on the pooled data from healthy subjects (7.0±3.6). In the case of the osteoporotic patients there was no significant difference between the predictive results using the predictor trained on pooled healthy data (3.5±1.5 time steps) and individually trained (2.3±0.8 time steps). However using the criterion of absolute predictability for each time series the normal group (3.9±1.2 prediction steps) was significantly ($P < 0.02$) further predictable into the future than the osteoporotic group (2.3±0.8 prediction; Table IV).

Fitting the best predictive ARIMA model (AR = 8, I = 1, MA = 9) to the pooled data from three healthy subjects and testing the predictability of this model on the remaining time series, we could separate the normal group by a significantly ($P < 0.02$) higher predictability (4.7±1.3 prediction steps) compared with the osteoporotic group (2.7±0.8 prediction steps; Table V). There was no significant difference in predictability compared to the neural network approach trained on pooled healthy data.

In addition, individual ARIMA models of different orders (AR-order: 1 . . . 15; I: 1 . . . 2; MA; 1 . . . 15) were fitted to each time series of normal subjects and osteoporotic patients. Using this procedure both groups were significantly ($P < 0.05$) further predictable into the future than using the individual neural network approach. However, due to a large variability in the degree of predictability the normal group (8.6±8.2 prediction steps) could not be separated significantly from the osteoporotic group (4.3±2.2 prediction steps). Comparing the best predictive ARIMA model for each time series with the results obtained from the network trained on pooled healthy data we could not find significant differences within the normal as well as the osteoporotic group.

Using pooled data from the osteoporotic patients and the "leave one out technique" for training, the healthy subjects

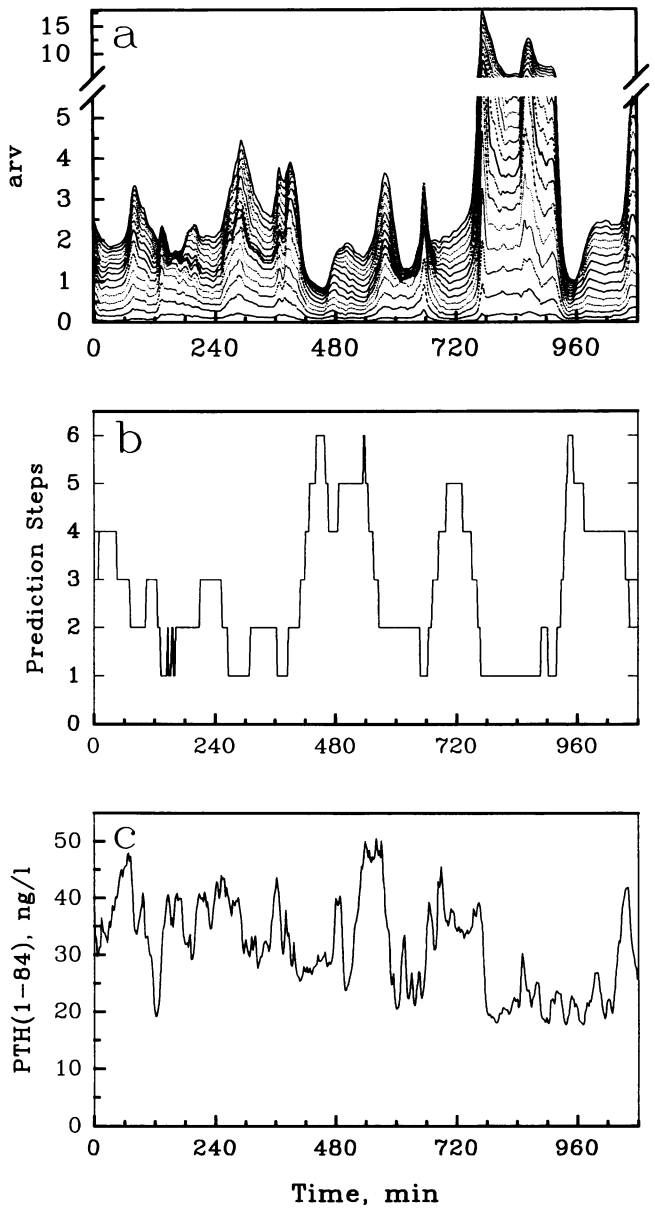

Figure 5. Nonuniformity of predictability in a single healthy subject. (*a*) arv (prediction error) vs. time for a 60 points (2 h) continuously running time window in a 21-h PTH serum concentration time series (acausally filtered) for 1–15 time steps ahead prediction (curves are from *bottom* to *top*). (*b*) Prediction step where the arv reached a value of 0.5 vs. time for a 60 points (2 h) continuously running time window. (*c*) Corresponding 21-h acausally filtered PTH serum concentration time series.

(6.1±2.9 prediction steps) were not significantly further predictable than the osteoporotic patients (4.8±3.3 prediction steps).

The healthy subjects were significantly more predictable than the osteoporotic patients using a variety of different forms of predictors and using predictors trained using both pooled data and on individual time series. Although the absolute difference in predictability varied with different prediction techniques, the consistency of the higher predictability for healthy subjects using a variety of techniques suggests that this higher predictability is due to a difference in the dynamics of the time

Table IV. Number of Prediction Steps (2 min) Until the Prediction Error (arv) Reaches a Value of 0.5

Subject	Prediction step
Normal1	6
Normal2	3
Normal3	4
Normal4	4
Normal5	5
Normal6	2
Normal7	4
Normal8	3
Normal9	4
Osteoporotic1	2
Osteoporotic2	2
Osteoporotic3	4
Osteoporotic4	2
Osteoporotic5	2
Osteoporotic6	2

The linear 15 input–1 output neural network was trained individually on the first two thirds of each time series.

Table V. Number of Prediction Steps (2 min) Until the Prediction Error (arv) Reaches a Value of 0.5

Subject	Prediction step
Normal1	6
Normal2	4
Normal3	6
Normal4	5
Normal5	5
Normal6	4
Normal7	6
Normal8	4
Normal9	2
Osteoporotic1	3
Osteoporotic2	2
Osteoporotic3	3
Osteoporotic4	2
Osteoporotic5	4
Osteoporotic6	2

The ARIMA model predictor was trained on pooled data from healthy subjects.

series for healthy subjects and is not merely the effect of a particular predictive model.

Taking only every 2nd data point for time series prediction, groups exhibiting low and high predictability as defined above could still be found but there was only a very small difference in the divergence rates. If the PTH concentrations were measured at 10-min intervals, it was impossible to make this distinction at all. This implies that secretory patterns evolve on a minute-to-minute time scale whereas their significance may only be seen in a profile over several hours.

Discussion

Two dynamic states? A hypothesis consistent with the results of our analysis is that PTH secretion in healthy subjects switches between a dynamic state of high predictability and a dynamic state of low predictability. The state of low predictability is characterized by a rapid divergence of the predicted and actual time series and a linear increase of arv with the number of prediction steps. This behavior is similar to the dynamics of the colored noise process, suggesting that the state of low predictability may be a low-order stochastic (random) state. The state of high predictability is characterized by less divergence between the predicted and actual time series and a saturation of the arv as the number of time steps is increased. Since a single network could predict equally well across many healthy subjects, it would appear that this deterministic component is common to all of the healthy subjects during some periods.

We found no evidence of a highly predictable saturating component in any of the osteoporotic time series using the approach with a neural network trained on pooled data from healthy subjects as well as with an individual neural network predictor derived from the patients themselves. This suggests that osteoporotic patients have lost the ability to switch from their dynamic state of low predictability to the dynamic state of high predictability.

Using the best predictive ARIMA model fitted on pooled healthy data, we could significantly separate both groups in analogy to the neural network approach, although the predictability was less than obtained with the neural network predictor.

The best predictive ARIMA model, individually fitted on each time series, performed better than the neural network approaches where the time series were trained and tested individually. The better prediction by the individual ARIMA models may be due to some nonstationarities in the data. However, the ARIMA models did not perform better than the neural network predictor trained on the pooled PTH data sets from healthy subjects. In particular, the ARIMA models could not separate the groups due to high variability in the predictive results.

The bimodal distribution of the predictability of the short time series from the nine healthy subjects (Fig. 4 *b*, Table III) may be due to the limited measurement period for these time series (between 4.5 and 9 h). Four of the healthy subjects have time series dominated by the state of high predictability, whereas the other five have time series dominated by the state of low predictability. The identification of distinct regions with low and high predictability in all of the longer time series from healthy subjects supports this hypothesis. Because periods of low predictability lasted between 2 h 24 min and 4 h 55 min in the long time series, it would not be surprising to find that the distributions of states of low and high predictability was not uniform in the short time series.

Our analysis, and reexamination of the longer series, suggests that PTH secretion seems to exhibit two distinct dynamic states. A model in which PTH secretion in healthy subjects switches between a dynamic state of high predictability and one of low predictability on an hourly to daily temporal scale would be biologically plausible given the temporal scale of the refractory period of the PTH receptor, its reduced affinity after hormone exposure (28) and the bone remodeling process with alternating phases of bone resorption and bone formation. Further evidence for a nonuniformity of PTH secretion can be seen by examining the most effective treatment protocol for osteoporosis. Daily injections of PTH show a tremendous gain

in bone mass in animals (11–13) and humans (14, 15), whereas continuous PTH infusion results in a loss of bone mass and structure.

The effectiveness of a daily PTH injection has been so surprising that it took a decade of consistent findings before becoming generally accepted. It is, however, not known whether this bone anabolic property of PTH finds a correlate in certain patterns of PTH secretion. Our data provide the first evidence that states of high predictability might be a regular finding in subjects with normal bone mass and metabolism. On the other hand the well-known minute-to-minute regulation of serum calcium could be of dominant influence.

If a high rate of change of PTH concentration is the significant factor in maintaining normal bone remodeling, more frequent application of PTH pulses with large amplitude may be even more effective than a daily injection. Application of large pulses of PTH on an hourly scale might be feasable with the use of a hormone pump. This is an important area for future investigation.

Two different temporal scales might be involved in the regulation of the bone remodeling process. One time scale, of ~ 2 min, contains the high-frequency fluctuations of PTH serum concentration, which may prevent the PTH receptors from down-regulating. The other scale is hourly to daily, where switching between states of low and high predictability is observed, which might be a "switch" between bone resorption and bone formation in the physiological bone remodeling process. The absence of switching to the highly predictable secretory state in osteoporotic subjects may then lead to the loss of bone mass and structure due to an imbalance of bone formation and bone resorption.

Nonuniform dynamics in other systems. Our suggestion of nonuniform (two state) dynamics in PTH secretion of healthy subjects is similar to that of Gallez and Babloyantz (29), who found evidence for switching between two different dynamic states in EEG data. Their study focused on variation of predictability in nonuniform attractors by investigating time series of EEG data. Recently Pawelzik et al. (30) demonstrated switching between predictable and unpredictable states in data from cat visual cortex. Gallez and Babloyantz proposed that if there is evidence for nonuniformity in the dynamics of attractors, it would be worth characterizing the predictability of parts of the attractor. The evidence that two regions of predictability exist in the PTH secretion, in EEG data, and in data from cat visual cortex suggests that it might be useful to investigate segments of other physiological signals such as cardiac rhythms or electrical activity of single neurons for distinct regions of predictability that may correspond to differing dynamic states. A similar approach has been used by Drepper (31) who calculated the predictability (using an information production profile) for a epidemiological time series of measles cases data. He also found nonuniformity in his attractor reconstructed from the measles cases data.

Differences in linear and nonlinear prediction. We found no significant difference in the predictive ability of linear and nonlinear networks for the PTH concentration time series. However, using classical nonlinear analysis, we found evidence for nonlinear determinism and possibly low-dimensional deterministic chaos in the irregular pattern of PTH secretion in healthy human subjects (32). Our results are in accordance with the findings of Blinowska and Malinowski (21) who demonstrated that the irregular nonlinear EEG signal could be predicted equally well or even better by a linear autoregressive model than by the nonlinear prediction model proposed by Sugihara and May (19). These results differ from others who compared linear and nonlinear predictive models, and this may be due to the relatively small amount of data available in our study, but a final clear conclusion is still lacking (19, 22, 24).

Although beyond the scope of this manuscript, an important area for future work is whether there is a nonlinear explanation of at least part of the behavior of PTH secretion. It may be possible to address this issue through the use of surrogate data (33). Surrogate data are simulated noisy time series that preserve certain statistical and dynamic features of the original time series from which they are generated. We are currently working on this type of analysis applied to the PTH time series.

A PTH biosensor is needed. Because switching between dynamic states of PTH secretion in healthy subjects seems to occur on a temporal scale of many hours, only extended measurements over periods of 1 d or longer could confirm our hypothesis of the nonuniformity of the PTH secretory dynamics. Unfortunately, the period over which PTH concentration can be measured frequently by blood sampling is severely limited by the resulting total blood loss.

It might be feasible to extend the measurement period to 2 d by reducing the sampling frequency from one blood sample every 2 min to one sample every 4 min. Longer measurement periods at such a high sampling frequency could only be performed by an on-line biosensor for PTH. Such a biosensor is not yet available although biosensors containing biological receptors such as the nicotinic acetylcholine receptor (34–36) and the L-glutamate receptor (37) have been developed. There is some hope of developing an on-line PTH-biosensor in the future because the PTH receptor has been cloned recently (38).

A sampling interval longer than 4 min to extend the measurement period is not recommended because separation into groups of low and high predictability is lost, as seen in our results with subsampling of the 2-min time series. This provides further evidence that biological information that separates the PTH secretory dynamics of healthy from osteoporotic subjects is encoded in the high-frequency fluctuations of the PTH serum concentration.

Time series prediction could be used for analyzing other dynamical diseases where methods such as computing the mean value for a time series or the power spectrum fail to distinguish normal from abnormal patterns. In particular, in such cases where a nonuniformity in the dynamics of a physiological attractor can be assumed, local time series prediction is a useful tool for characterizing different dynamic states. Our results suggest that the PTH secretory pattern in healthy subjects is an example of nonuniform dynamics that exhibit at least two different phases of secretion, one dynamic phase of high predictability and one of low predictability.

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References

1. Harms, H. M., U. Kaptaina, W. R. Külpmann, G. Brabant, and R. D. Hesch. 1989. Pulse amplitude and frequency modulation of parathyroid hormone in plasma. *J. Clin. Endocrinol. Metab.* 69:843–851.

2. Kitamura, N., C. Shigeno, K. Shiomi, K. Lee, S. Ohta, T. Sone, S. Katsushima, E. Tadamura, T. Kousaka, and I. Yamamoto. 1990. Episodic fluctuation in serum intact parathyroid hormone concentration in men. *J. Clin. Endocrinol. Metab.* 70:252–263.

3. Parthemore, J. G., B. A. Ross, D. C. Parker, D. F. Kripke, L. V. Avioli, and L. J. Deftos. 1978. Assessment of acute and chronic changes in parathyroid hormone secretion by a radioimmunoassay with predominant specificity for the carboxy-terminal region of the molecule. *J. Clin. Endocrinol. Metab.* 47:284–289.

4. Kripke, D. F., P. Lavie, D. Parker, L. Huey, and L. F. Deftos. 1978. Plasma parathyroid hormone and calcium are related to sleep stage cycles. *J. Clin. Endocrinol. Metab.* 47:1021–1027.

5. Howard, G. A., B. L. Bottemiller, and D. Baylink. 1981. Parathyroid hormone stimulates bone formation and resorption in organ culture: evidence for a coupling mechanism. *Proc. Natl. Acad. Sci. USA.* 78:3204–3208.

6. Huffer, W. E. 1988. Morphology and biochemistry of bone remodeling: possible control by vitamin D, parathyroid hormone, and other substances. *Lab. Invest.* 59:418–442.

7. McSheehy, P. M. J., and T. J. Chambers. 1986. Osteoblast-like cells in the presence of parathyroid hormone release soluble factor that stimulates osteoclastic bone resorption. *Endocrinology.* 119:1654–1659.

8. Mundy, G. R. 1989. Local factors in bone remodeling. *Recent Prog. Horm. Res.* 45:507–527.

9. Raisz, L. G. 1988. Local and systemic factors in the pathogenesis of osteoporosis. *N. Engl. J. Med.* 318:818–828.

10. Wong, G. L. 1984. Paracrine interactions in bone-secreted products of osteoblasts permit osteoclasts to respond to parathyroid hormone. *J. Biol. Chem.* 259:4019–4022.

11. Liu, C. C., D. N. Kalu, E. Salerno, R. Echon, B. W. Hollis, and M. Ray. 1991. Preexisting bone loss associated with ovariectomy in rats is reversed by parathyroid hormone. *J. Bone Miner. Res.* 6:1071–1080.

12. Podbesek, R., C. Edouard, P. J. Meunier, J. A. Parsons, J. Reeve, R. W. Stevenson, and J. M. Zanelli. 1983. Effects of two regimes with synthetic human parathyroid hormone fragment on bone formation and the tissue balance of trabecular bone in greyhounds. *Endocrinology.* 112:1000–1006.

13. Tam, C. S., J. N. Heersche, T. M. Murray, and J. A. Parsons. 1982. Parathyroid hormone stimulates the bone apposition rate independently of its resorptive action: differential effects of intermittent and continuous administration. *Endocrinology.* 110:506–512.

14. Hesch, R. D., U. Busch, M. Prokop, G. Delling, and E. F. Rittinghaus. 1989. Increase of vertebral density by combination therapy with pulsatile 1–38hPTH and sequential addition of calcitoninn nasal spray in osteoporotic patients. *Calcif. Tissue Int.* 44:176–180.

15. Slovik, D. M., D. I. Rosenthal, S. H. Doppelt, J. T. Potts, M. A. Daly, J. A. Campell, and R. M. Neer. 1986. Restoration of spinal bone in osteoporotic men by treatment with human parathyroid hormone (1-34) and 1,25 dihydroxyvitamin D. *J. Bone Miner. Res.* 1:377–381.

16. Mackey, M. C., and L. Glass. 1977. Oscillation and chaos in physiological control systems. *Science (Wash. DC).* 197:287–289.

17. Mackey, M. C., and J. G. Milton. 1987. Dynamical diseases. *Ann. NY Acad. Sci.* 504:16–32.

18. Casdagli, M., D. Des Jardins, S. Eubank, J. D. Farmer, J. Gibson, N. Hunter, and J. Theiler. 1991. Nonlinear Modeling of Chaotic Time Series: Theory and Applications. Los Alamos National Laboratory, Los Alamos, NM. (Technical Report No. LA-UR-91-1637)

19. Sugihara, G., and R. M. May. 1990. Nonlinear forecasting as a way of distinguishing chaos from measurement error in time series. *Nature (Lond.).* 344:734–741.

20. Tsonis, A. A., and J. B. Elsner. 1992. Nonlinear prediction as a way of distinguishing chaos from random fractal sequences. *Nature (Lond.).* 358:217–220.

21. Blinowska, K. J., and M. Malinowski. 1991. Non-linear and linear forecasting of the EEG time series *Biol. Cybern.* 66:159–165.

22. Lapedes, A. S., and R. M. Farber. 1987. Nonlinear signal processing using neural networks: prediction and system modeling. Los Alamos National Laboratory, Los Alamos, NM. (Technical Report No. LA-UR-87-2662)

23. Nowlan, S. J., and G. E. Hinton. 1992. Simplifying neural networks by soft weight-sharing. *Neural Comput.* 4:473–493.

24. Weigend, A. S., B. A. Huberman, and D. E. Rumelhart. 1990. Predicting the future: a connectionist approach. *Int. J. Neural Syst.* 1:193–209.

25. Chatfield, C. 1984. The Analysis of Time Series. 3rd ed. Chapman and Hall, London.

26. Pincus, S. M., and D. L. Keefe. 1992. Quantification of hormone pulsatility via an approximate entropy algorithm. *Am. J. Physiol.* 262:741–754.

27. Mitschke, F. 1990. Acausal filters for chaotic signals. *Phys. Rev. A* 41:1169–1171.

28. Pun, K. K., P. W. M. Ho, R. A. Nissenson, and C. D. Arnaud. 1990. Desensitization of parathyroid hormone receptors on cultured bone cells. *J. Bone Miner. Res.* 5:1193–1200.

29. Gallez, D., and A. Babloyantz. 1991. Lyapunov exponents for nonuniform attractors. *Phys. Lett.* 161:247–254.

30. Pawelzik, K., H. U. Bauer, and T. Geisel. 1993. Switching between predictable and unpredictable states in data from cat visual cortex. *In* Proceedings of CNS 92 San Francisco. F. H. Eeckman and J. Bower, editors. Kluwer Academic, Dordrecht.

31. Drepper, F. 1988. Unstable determinism in the information production profile of an epidemiological time series. *In* Ecodynamics. W. Wolff, C. J. Soeder, and F. Drepper, editors. Springer-Verlag, Berlin. 319–332.

32. Prank, K. H. Harms, M. Dämmig, G. Brabant, F. Mitschke, and R. D. Hesch. 1994. Is there low-dimensional chaos in pulsatile secretion of parathyroid hormone in normal human subjects? *Am. J. Physiol.* 266:E653–E658.

33. Theiler, J. B. Galdrikan, A. Longtin, S. Eubank, and J. D. Farmer. 1992. Using surrogate data to detect nonlinearity in time series. *In* Nonlinear Modeling and forecasting. SFI Studies in the Sciences of Complexity. Proc. Vol. XII. M. Casdagli and S. Eubank, editors. Addison-Wesley, Redwood City. 163–188.

34. Eldefrawi, M. E., S. M. Sherby, A. G. Andreou, N. A. Mansour, Z. Annau, N. A. Blum, and J. J. Valdes. 1988. Acetylcholine receptor-based biosensor. *Anal. Lett.* 21:1665–1680.

35. Gotoh, M., E. Tamiya, M. Momoi, Y. Kagawa, and I. Karube. 1987. Acetylcholine sensor based on ion sensitive field effect transistor and acetylcholine receptor. *Anal. Lett.* 20:857–870.

36. Gotoh, M., E. Tamiya, and I. Karube. 1989. Micro-FET Biosensors using polyvinylbutyral membrane. *J. Membrane Sci.* 41:291–303.

37. Uto, M., E. K. Michaelis, I. F. Hu, Y. Umezawa, and T. Kuwana. 1990. Biosensor development with a glutamate receptor ion-channel reconstituted in a lipid bilayer. *Anal. Sci.* 6:221–225.

38. Jüppner, H., A. B. Abou-Samra, M. Freeman, X. F. Kong, E. Schipani, J. Richards, L. F. Kolakowski, Jr., J. Hock, J. T. Potts, Jr., and H. M. Kronenberg. 1991. A G-protein linked receptor for parathyroid hormone and parathyroid hormone related peptide. *Science (Wash. DC).* 254:1024–1026.