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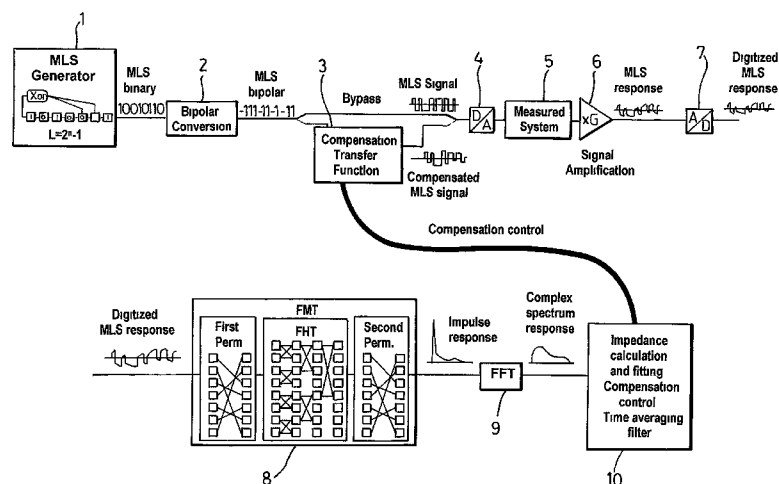
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(54) **Title:** MEASUREMENT OF THE ELECTRICAL IMPEDANCE FREQUENCY SPECTRUM



(57) **Abstract:** The invention provides apparatus for measuring the AC electrical parameters of a sample, such as an electric component, electric network, material when dry or in liquid, the apparatus comprising a generator for generating a pseudo random binary sequence, a digital to analogue (D/A) unit or equivalent, or other means to output the sequence in digital or analogue format, first connection means for connecting the output of the generator to the input of the D/A, sample input connection means adapted to apply the output of the D/A to the sample, sample output connection means for connecting a sample output to an analogue to digital converter (A/D) or equivalent, and an output processing unit being configured for subjecting the output of the A/D to a Hadamard Transform followed by a Fast Fourier Transform (FFT) for producing in use a measurement of the complex impedance frequency response of the sample. The inventive apparatus can acquire the impedance frequency spectrum (calculated from the measured transfer function) of a system in a very short time, typically 1 microsecond to 5 seconds.

MEASUREMENT OF THE ELECTRICAL IMPEDANCE FREQUENCY SPECTRUM

Field of the Invention

This invention relates to methods and apparatus for measuring the
5 electrical impedance frequency spectrum of devices or samples, etc.

The invention relates particularly, but not exclusively, to methods and
apparatus for measuring the electrical impedance spectrum of a device or
array of devices, such as an electric component, electric network,
material or sample when dry or in a liquid, preferably with provision for
10 recording the impedance frequency spectrum as a function of time.
Electrical signals applied to the object being inspected may be subject to
a compensation transfer function.

For convenience, the term 'sample' has been used herein to refer where
appropriate to any such device, array of devices, material/s, sample,
15 tissue, organ or biological entity, or combination thereof, in vitro or in
vivo, being measured.

Summaries of the Invention

According to one aspect of the invention we provide a method for
measuring the AC electrical parameters of a sample, as hereinbefore
20 defined, comprising generating a pseudo random binary sequence and
using the sequence to control a voltage or current electrical signal
applied to the sample, using a digital to analogue converter (D/A) or
equivalent, or the digital signal itself, measuring the response of the
sample to said electrical signal by subjecting the output from the sample
25 to an analogue to digital converter (A/D) or equivalent, and processing

the output of the A/D with a Hadamard Transform in conjunction with a Fast Fourier Transform (FFT) to produce a measurement of the complex impedance frequency response of the sample.

Preferably the pseudo random binary sequence is modified by a
5 compensation transfer function before being applied to the sample, the compensation transfer function having been configured to compensate for at least some unwanted response component of the sample under test.

According to another aspect of the invention we provide apparatus for measuring the AC electrical parameters of a sample, the apparatus
10 comprising a generator for generating a pseudo random binary sequence, a digital to analogue [D/A] unit or equivalent, or other means to output the sequence in digital or analogue format, first connection means for connecting the output of the generator to the input of the D/A, sample input connection means adapted to apply the output of the D/A to the
15 sample, sample output connection means for connecting a sample output to an analogue to digital converter (A/D) or equivalent, and an output processing unit being configured for subjecting the output of the A/D to a Hadamard Transform followed by a Fast Fourier Transform (FFT) for producing in use a measurement of the complex impedance frequency
20 response of the sample.

Embodiments of the present invention relate to the acquisition of the impedance frequency spectrum (calculated from the measured transfer function) of a system in a very short time, typically 1 microsecond to 5 seconds. Such a method is faster than traditionally used sweep frequency
25 techniques because the signal used to interrogate the system has a broad spectrum, thus interrogating the system simultaneously over the complete spectrum of interest. The signal can be processed with minimal time delay. The signal applied to the system is subject to compensation to

cancel noise from other non-correlated noise sources. The signal processing uses a Hadamard Transform to compute the impulse response of the system. The compensation will generally comprise both amplitude and phase compensation applied to the signal prior to its use to
5 interrogate the system. This is done using a transfer function on the signal so configured as to compensate a posteriori (with previous knowledge) for some or all, measured, known or chosen properties of the system spectrum or for parasitic effects.

In various embodiments the device or system being measured is an
10 electric component, an electric network or circuit, a material organic or inorganic, a biological tissue, organ or part of the human body in vivo or in vitro, biological cell or cells or other particles, biological particles or powders, dry or wet, in liquid, gas, or vacuum. The device or system is connected to the instrument through wires and contacts or electrodes,
15 preferably microelectrodes. The electrical impedance of these contacts, electrodes or wires can affect the measured impedance over a wide spectrum. These effects or artefacts can be measured as part of the system response in a setting up procedure and can be considered to be unwanted parasitic effects which somehow alter the measurement of the
20 actual device or system. The transfer function can then be configured on the basis of the measurements made in the setting up procedure and/or on the basis of knowledge of the system being tested to compensate for these artefactual or parasitic effects such as, but not limited to, electrode polarization and can thereby improve the sensitivity of the measurement.

25 Compensation can also provide the means to alleviate some of the problems related to the dynamic range due to the A/D converter or amplifier electronics.

Background to the Invention

In the prior art it was shown that a pseudo-random binary sequence could be used [1, 2] to interrogate materials for gas sensing. The authors of the article and patent application use a coherent demodulation and an FFT
5 (Fast Fourier Transform) to perform the signal processing. No compensation is made to the signal being applied to the system or to the response. Their system is able to detect a change in material properties, as some spectrum signature, but is not sufficient to obtain a complex spectrum impedance data of the sample. Another important difference is
10 that the spectrum response is passed through a coherent demodulator which selects a single frequency of interest at a time. Thus, a complete frequency spectrum is not measured for each sequence recorded by the system. A complete spectrum is thus not obtained for each sequence repetition. The coherent demodulator does not use knowledge of the
15 applied sequence to suppress noise from uncorrelated noise all over the spectrum. The uncompensated technique which has an associated low dynamic range is also not suitable if the measurement has to be carried over a few decades in frequency simultaneously on systems which comprise a frequency dependent equivalent impedance element (such as a
20 capacitor like element).

A number of post-processing methods have been previously reported [3] to correct for electrode polarization effects on the measured data with more or less success. Yet, none of them do compensate by adapting the frequency spectrum of the signal being applied to the system.

25 Various embodiments of the invention will be described hereafter, by way of example only, with reference to the accompanying drawings.

Brief Description of the Figures

In the drawings:

5 **Figure 1a** is a block diagram showing, in the upper part thereof, apparatus for providing a compensated MLS signal to a D/A connected to the system to be measured, and an A/D connected to the system output to provide a digitised output, and in the lower part thereof output signal processing means for providing a complex spectrum response. In addition an impedance calculation unit is shown for providing compensation to control the compensation transfer function,

10

Figure 1b shows the detail of the measured system in Figure 1a for the typical case of two electrodes used to measure a liquid including particles in suspension or some contaminants,

15 **Figure 2** shows a system for differential measurement which can be used with two or more electrodes associated with the sample under test,

Figure 3 shows a system for multiple measurements in a multiple input, multiple output system,

20 **Figure 4** shows in the upper half a system suitable for an array, and in the lower half a modification to that system suitable, *inter alia*, for impedance tomography,

Figure 5 shows schematically an application of the invention to impedance flow cytometry,

Figure 6 shows a spectrogram of a time varying system measured using the apparatus of Figure 5,

Figure 7 schematically shows application of the invention to a cell positioned on top of one of the electrodes,

5 **Figure 8** shows measurements being made of a cell located over a hole with electrodes placed on each side of the hole, and

Figures 9 and 10 show the application of the invention to the measurement of monolayers which may contain membrane proteins.

With reference to Figure 1a, a binary maximum length sequence MLS is
10 generated in unit 1 using a circular shift register of order n. Unit 2 is an optional unit arranged to convert the binary sequence produced by 1 to a bipolar sequence replacing all zeros of the original sequence by ones and all ones by minus 1. Use of a bipolar sequence is preferred so as to reduce the signal offset.

15 The bipolar or binary sequence is passed in normal use through a filter 3 which is preferably a digital filter of the (finite impulse response) FIR type acting as a compensation transfer function. Bypass switches are provided to enable unit 3 to be bypassed so as to generate, when required, a flat signal spectrum in the cases when the response spectrum
20 of the sample is relatively flat or when knowledge of an unknown system is being acquired. The signal is converted to an electric signal using a digital to analogue (D/A) converter 4. The clock frequency of the conversion performed by D/A 4 determines the frequencies at which the system is measured. The amplitude of the analogue signal can be
25 additionally controlled at this stage. The signal from the output of the

D/A 4 is passed through the system 5 to be measured. The systems include all wires and connection to the device of interest.

Figure 1b shows the electrodes used to measure the spectrum response of a liquid including particles.

5 The system can be a time variant system.

An optional signal amplification stage 6 can be used to adapt the output response of system 5 for the subsequent digitalization. The response is digitized by A/D 7 in order to be further processed.

10 In module 8 the digitized response from the output of A/D 7 is processed using a Fast-M-Transform (FMT) which comprises a Hadamard Transform or Fast Hadamard Transform to obtain the impulse response of the system. The impulse response of the system is processed using a FFT 9 to obtain the complex spectrum response of the system. The complex spectrum response is converted in block 10 to the impedance
15 spectrum of the system.

The filter parameters for the compensation transfer function 3 can be calculated in a setting up procedure in which the bypass to unit 3 is employed. A time averaging filter can be used at this stage to suppress slow variation of offset in the impedance spectrum if one wants to
20 recover the variance in time of the system.

Pseudo random binary sequence used for impedance measurement

The measurements in Figure 1a use a Pseudo-Random Binary Sequence (PRBS) generated in unit 1, preferably a Maximum Length Sequence (MLS) which can be logically generated using an implementation of a

shift register or by software [4]. The order of the shift register n is related to the length of the sequence / according to the equation $L = 2^n - 1$.

The order of the shift register is typically between 4 and 16. The length of the sequence is thus typically between 15 and 65535. The logical
5 binary sequence is preferably converted to a numerical bipolar sequence of selected amplitude. The sequence is then converted to a voltage or current electrical signal using the digital-to-analogue D/A converter 4 and applied to the device 5, material or sample using one, two or more excitation contacts or electrodes as will be described hereafter. A number
10 of contact or electrodes are used to obtain the response from the system.

Instead of a D/A converter 4, the logical signal can also be applied to the device 5 using an operational amplifier circuit, or a transistor, which produces an analogue signal with the same pattern and frequency as the digital signal but with a defined selectable amplitude and offset. The
15 power spectrum of the generated signal is flat up to the Nyquist frequency (which is half the frequency of the clock frequency used in the D/A converter) .

The Hadamard transform of unit 8 is used to compute the impulse response from the system response. Its computation is further described
20 by people knowledgeable in room acoustic measurements [5] .

Fast **Impedance Spectrogram**

Because the whole spectrum can be measured each time a sequence is sent to the system a spectrogram of the system complex transfer function or derived complex impedance can be measured a few thousand times per
25 second or more using this method. This is important if rapid changes in the system impedance spectrum are significant for the measurement.

Frequency dependent compensation (transfer function)

The invention is concerned with compensating at least in part for the impedance spectrum of the system being measured (Figure 1a). This compensation uses a transfer function which is applied to the binary
5 sequence or a derived sequence to compensate for a frequency dependent behaviour of the measured system, measuring electronics, contacts or contacts leads. It was found advantageous to place this compensation before the system so as to modify the signal being applied and obtain significant signal amplitude for frequencies where the system impedance
10 is large, rather than applying a signal having a flat frequency spectrum, and then trying to compensate for small response amplitude of the system at certain frequencies.

The generated bipolar numerical sequence or a derived sequence can be passed through a transfer function which compensates a posteriori for
15 certain measured or known electrical parameters or transfer function of the device, material or sample. The compensation can be bypassed when required to measure the uncompensated system. In the case of a transfer function differing from a simple gain, the waveform generated using the digital-to-analogue converter no longer has a flat power spectrum.

20 As an example, in the case of electrodes in a liquid the transfer function can be made to compensate for the frequency behaviour of the electrical double layer polarization which usually takes the form of a capacitor or a constant phase element. The interfacial impedance generated by the double layer can be very large at low frequencies compared with the rest
25 of the system and thus can screen the measurement at those frequencies.

Compensating for such an interfacial impedance in series with some resistive elements of interest can be done by using a transfer function

which proportionally increases the voltage of the low frequency components of the sequence. In the case of a bipolar voltage applied to a device material or sample using two electrodes, this compensates for the smaller signal amplitude measured at the lower end of the frequency spectrum where the voltage drop through the series resistive elements is
5 small compared to the voltage drop through the interfacial capacitance.

The compensation transfer function can also compensate for, but not limited to, parasitic stray capacitors, series resistors or other electrical elements either in parallel or series with or inherent to the device,
10 material or sample being measured. These parasitic effects can have an influence on a part or the whole measured spectrum, the compensation transfer function can thus be used to compensate for those effects by modifying the spectrum of the applied sequence so as to cancel their effect on the measured spectrum.

15 If necessary, a linear, square or logarithmic, or other non-linear amplification stage can be used to scale the output amplitude of the digital-to-analogue converter.

The uncompensated measured spectrum can be used to select the parameters of the compensation transfer functions, or be used directly for
20 measurement when the spectrum over the frequency range of interest is found to be relatively flat.

The compensation transfer function can also be used to compensate for distortions in the spectrum introduced by an analog-to-digital or a digital-to-analogue converter, by analogue filters, or by one or more signal
25 amplification stages. Any unwanted frequency dependent or independent phase effect can also be compensated by the transfer function.

Interface to the device and differential measurement

A set of one, two or more electrodes can be used to make electrical contact to the device, material or sample being measured. Optionally, a differential measurement using a second set of one, two or more
5 electrodes applied to a different part of the same or to a different reference device, material or sample can be used simultaneously or at a different time.

Advantage of this invention to simplify prior art based instruments and achieve more accurate measurement:

10 In prior art instruments (typically called impedance analyzers) the user generally chooses a fixed voltage which is applied throughout the whole frequency sweep of the spectrum. If the equivalent of a frequency-dependent impedance element is present in the measured system, the measured response amplitude, in this case a current, can vary over the
15 frequency spectrum by many order of magnitude. This consequently requires adapting the gain of the amplification stage of the instruments as a function of the measured frequency range in order to avoid saturation or achieve a required resolution.

In this invention, because all of the measured frequencies are sent
20 simultaneously, within one sequence, through the system, adapting the amplifiers gain as a function of time throughout a sweep is not possible. Modifying the synthesized flat broad spectrum sequence being generated to compensate for the system measured or known transfer function using the compensation transfer function enables selected parts of the applied
25 spectrum to be changed up or down in magnitude or phase.

This compensation method does amongst other advantages suppress the need to adapt the amplification stage gain as a function of frequency and thus enables to simplify its embodiment as an instrument compared to prior art, while offering a good dynamic range over the measured spectrum.

The compensation transfer function can be adapted in real-time by a computer, a DSP or other means to compensate for drift in the system, such as changes in the interface between electrodes and liquid.

One implementation of this compensation transfer function can be a Finite Impulse Response (FIR) filter with delays and gains as tunable parameters. An alternate implementation would be an Infinite Impulse response (IIR) filter.

It would be desirable in some cases to be able to arrange for some compensation is made right after the system or in the digital processing of the response. This method is of interest only if the signal applied to the system needs to be of even amplitude over the whole frequency range, for instance to avoid a non-linearity in the response, for a particular measurement requirement.

Differential measurement

In Figure 2 a compensation transfer function assembly comprises two functions 1 and 2. Compensation transfer function 1 is used in the reference branch of the differential measurement. It can be adapted in real-time by the computer to compensate for common effects found in the two branches. Compensation transfer function 2 is used in the measured branch of the differential measurement and can compensate for differences between the measured branch and the reference branch. The

system of Figure 2 may contain additional differentially measured branches according to the system to be measured.

The compensation transfer functions 1 and 2 of unit 1 can be adapted in real-time by a computer, a DSP or other means to compensate for drift in
5 the system, such as changes in the interface between electrodes and liquid, common mode and differences between the different branches. The sequences for the reference and measured branches are passed through separate digital-to-analogue converters 12, and the signals are applied to the same system 13 or to a related system. The signals output
10 from the system 13 are differentially amplified by amplifier 14 or somehow compared to the reference. The differential response of the system is converted by A/D 15 to a digital response and post processed.

In the case of the differential measurement (Figure 2) this can compensate for a common and for differential property of the device or
15 devices, materials or samples. For example, in the case of two or more electrodes in a liquid the differential-transfer function can be made to compensate for any difference in interfacial polarization found between the electrodes. In the case of a material the transfer function can compensate for a difference in contact impedance or in the impedance of
20 the contact leads. Compensating for the common properties can be done by measuring each branch independently prior to measuring them differentially.

Multiple measurement

In the case of multiple measurements (Figure 3) the sequence can be
25 passed through two or more separate transfer functions to compensate for common mode and differences in the different elements of the system.

Compensation transfer functions of compensation transfer unit 16 1 to n are used for each individual branch of an array type of measurement. They can be adapted in real-time by a computer, a DSP or other to compensate for common mode and differences between the n different
5 branches. The signals for the different branches are converted through separate digital-analogue converters 17. The system 18 being measured can be an array of samples or a single sample measured with multiple electrodes. The multiple responses are optionally amplified individually by amplifiers 19. The multiple responses of the system or systems are
10 converted to a number of digital responses by A/D'S 20 and post processed using a computer or DSP,

The response from each element can be amplified, digitized and processed as individual channels. This can be considered a multiple input multiple output system.

15 A single channel can be used instead in the amplification with time multiplexing if the different elements of the array need not be measured simultaneously. This can be considered a multiplexed input - demultiplexed output system. In this case it is still possible to apply differential compensation for each measurement by changing the filter
20 transfer function synchronously with the time multiplexing for each element.

For array of sensor or electrical impedance tomography (Figure 4) types of applications it is possible to use a single sequence which is applied to a single system having multiple electrodes or contacts or an array of
25 sensors.

In Figure 4 compensation transfer function 21 is used to compensate for common mode or can be related to the electrode role. The sequence is

passed through a single digital to analogue converter 22. The system 23 being measured, as in the upper part of Figure 4, can be an array of samples sharing a common contact or electrode or with one or more contact or electrodes connected to the same sequence, or as shown in the lower part of Figure 4, the system 23 *bis* being measured can also be connected using contacts or electrodes whose respective role (excitation or measurement) can be exchanged in time. In the case illustrated in the modification of 23 *bis* there is one excitation contact or electrode and multiple measurement contact or electrodes. The response signals are optionally amplified by amplifiers 24. The multiple responses of the system or systems are converted to a number of digital responses by A/Ds 25 and post processed using a computer or DSP.

In the array of sensor, if only common (as opposed to differential) compensation is required, the same signal can be applied to all the sensors simultaneously. Using multiple detection channels allows for simultaneous recording of the impedance in the sensor but multiplexing can also be used.

In the impedance tomography case, the sequence is applied to 1 electrode the other electrodes are used to measure the different responses of the system the respective role of the electrodes can be changed in time so as to apply the sequence to the system using a different electrode and measure the response from all others. A different compensation can be used each time the role of the electrodes is exchanged.

Finding the compensation parameters

Uncompensated or partly compensated measurement of the impedance or differential impedance of the device, material or sample can be used to gain knowledge of the compensation transfer function. For instance, if

the inverse transfer function of a measured system (with the digital filter disabled) is then used in the compensation, the measured spectrum of the system will be made flat, provided the system is time invariant.

The transfer function parameters can also be adapted in real time through a feed-back loop to follow changes in the measured spectrum. This is typically done by using a time average filter on the measured spectrum with a time constant which is long compared to the change in frequency spectrum which is being monitored. This permits to adjust the parameters of the compensation transfer function while it is active by compensating for relatively long term change to the system impedance. Provisions are made to record the transfer function used in the digital filter compensation as it contains the slowly changing or time invariant properties of the device, material, sample as well as contact and wire information.

15 **Electrode configurations**

A number of electrode configurations are presented here but it will be appreciated that this is not intended to limit the number of possible configurations the system can measure using the disclosed method.

For a certain configuration of electrodes the sequence can be applied between two excitation electrodes either by applying the sequence to one electrode and keeping a fixed potential the other or by applying the sequence to one electrode and the phase opposed signal to the other. Two or more measurement electrodes are used to obtain the voltage drop between two or more points of the device, material or sample.

For another configuration of electrodes the sequence is applied to one or more electrodes and one or more measurement electrodes are connected

to a virtual ground. In this case, the current passing through the virtual ground is measured for each measurement electrode and is subsequently converted to a voltage using a trans-impedance amplification stage or by measuring the voltage drop across a resistor placed in series with the sample. In another configuration, the role of the electrodes can optionally be changed rapidly which enables performing fast time resolved impedance tomography. The compensation can be changed synchronously with the change of the roles of the electrode so that each possible electrode role configuration can use a different compensation.

10 Signal processing

The measured response is optionally amplified electronically. In certain configuration it can be amplified differentially.

It is digitized using an analogue to digital converter clocked synchronously to the digital to analogue converter at the same frequency, or at a divider of two of the digital to analogue converter frequency. The rest of the processing can be done using any type of computing means, a computer, a digital signal processor (DSP) or an integrated circuit. The digitized response data is organized into successive time frames comprising 1 digitized response samples. Each of those response frames corresponds in time to a generated sequence. This is possible as the converters work synchronously.

The digitized response is transformed into the impulse response of the device, material or sample using a Fast M Transform (FMT) comprising a Hadamard Transform or Fast Hadamard Transform (FHT).

Compared to state of the art methods which simply use FFT or short time FFT (STFFT), this technique is considerably more noise immune as the

signal processing uses knowledge of the signal applied to the system to remove noises from other non-correlated noise sources.

The FMT computation involves only a fraction of the FFT computation.

The impulse response is optionally padded with a zero before being
5 transformed into the complex frequency response using a Fast Fourier Transform.

Additionally time averaging can be performed if one is interested in changes in the measured spectrum, thus removing low frequency drifts and offsets in the measured spectrum.

10 Applications

A typical example is found in impedance spectroscopy flow cytometry [6-9] Figure 5 where the user is primarily interested in the rapid changes to the system due to the cell passing, and other time independent or slowly time dependent information are only secondary. A spectrogram of a time
15 varying system measured using the disclosed method is shown in Figure 6. In this case, a differential complex spectrum is measured for 512 different frequencies every millisecond. Only the real part of the spectrogram is shown, it is additionally corrected for offset using time averaging. The system comprises particles passing in a capillary channel
20 between a specific electrode arrangement according to the literature [6, 7].

Knowledge on the dielectric properties of passing particles or cells can be retrieved by looking at the change in time of the measured spectrum.

Additional processing such as time and spectrum filtering can be used. Single cell parameters extraction can be done using spectrum fitting algorithm or by matching the measured data to previously acquired data.

Additional applications including cell as the sample are by no mean
5 limited to but include measurement of cell on a surface or on top of one of the electrodes Figure 7. One or more cells over one or more holes with electrodes placed on each sides of the hole Figure 8.

In impedance tomography the ability to rapidly measure the impedance spectrum using multiple electrodes geometrically positioned relatively to
10 the sample and change the role of those electrodes (excitation or measurement) is important Figure 4. This technique enables fast time resolved impedance tomography. The compensation transfer function can be changed according to which electrode is used for excitation.

Other biological applications include the measurement of monolayers and
15 membranes which may additionally contain membrane proteins. These can be measured either on surfaces Figure 9 or on holes Figure 10.

Knowledge of the device, material or sample can be retrieved as a complex impedance spectrum using the complex frequency response of the system and the applied signal spectrum.

20 Electrical parameters of interest are obtained by further analyzing the complex impedance spectrum of the system using fitting procedures such as complex non-linear least square fitting or matching to a knowledge base previously acquired.

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CLAIMS

1. A method for measuring the AC electrical parameters of a sample, as hereinbefore defined, comprising generating a pseudo random binary sequence and using the sequence to control a voltage or current electrical
5 signal applied to the sample, using a digital to analogue converter (*DJA*) or equivalent, or the digital signal itself, measuring the response of the sample to said electrical signal by subjecting the output from the sample to an analogue to digital converter (*A/D*) or equivalent, and processing the output of the *A/D* with a Hadamard Transform in conjunction with a
10 Fast Fourier Transform (*FFT*) to produce a measurement of the complex impedance frequency response of the sample.
2. A method according to claim 1 in which the Hadamard Transfer Function is the Fast Hadamard Transfer Function.
3. The method of claim 1 or claim 2 in which the pseudo random
15 binary sequence is modified by a compensation transfer function before being applied to the sample, the compensation transfer function having been configured to compensate for at least some response component of the sample under test.
4. A method according to claim 3 in which the compensation transfer
20 function is configured utilising knowledge obtained by measuring the impedance frequency spectrum of the sample with the compensation transfer function being disabled.
5. A method according to any one of the preceding claims in which the compensation transfer function is implemented using a digital filter.

6. A method according to claim 5 in which said digital filter is a finite impulse response (FIR) filter.
7. A method according to claim 5 in which the said digital filter is an infinite impulse response (HR) filter.
- 5 8. A method according to any one of claims 3 to 7 in which the compensation transfer function is repeatedly adapted in time, with minimal or with a short delay after having acquired some knowledge of the system.
9. A method according to any one of the preceding claims in which the
10 compensation transfer function applies a linear, square or logarithmic, or other non-linear, amplification to scale the output of the digital to analogue converter supplying the electrical signal to the sample.
10. A method according to claim 9 in which the output amplitude of said digital to analogue converter scales over two orders of magnitude or
15 more.
11. A method according to claim 9 in which the output amplitude of the digital to analogue converter is logarithmic.
12. A method according to any one of the preceding claims in which the sample is interfaced using two or more electrodes or contacts.
- 20 13. A method according to claim 12 in which one or more sequences are multiplexed so that the sequence can be applied to the two or more electrodes or contacts sequentially in time.

14. A method according to claim 13 in which the measured response output from two or more electrodes or contacts are demultiplexed.
15. A method according to any one of the preceding claims in which the signal collection comprises a differential amplification stage using a second set of electrodes, or any number of additional electrodes, or contacts on the same sample or on a related sample.
16. A method according to claim 3 or any one of the preceding claims each as appended to claim 3 in which the said compensation transfer function is configured to compensate at least in part for electrode polarization or contact impedance to the sample.
17. A method according to claim 3 or any one of the preceding claims each as appended to claim 3 in which the said compensation transfer function is configured to compensate at least in part for non-ideal gain of one or more analogue amplifiers.
18. A method according to claim 3 or any one of the preceding claims each as appended to claim 3 in which the said compensation transfer function compensates at least in part for non-ideal behaviour of an analogue to digital or digital to analogue converter.
19. A method according to claim 3 or any one of the preceding claims each as appended to claim 3 in which the said compensation transfer function is configured to compensate at least in part for non-ideal contact leads.
20. A method according to claim 3 or any one of the preceding claims each as appended to claim 3 in which the said compensation transfer

function is configured to compensate at least in part for a stray capacitance.

21. A method according to any one of the preceding claims in which the processing of the output of the sample is done in a time efficient manner
5 so as to process continuously the data being digitized by the A/D in order to obtain for each sequence repetition a full spectrum of the response.

22. A method according to any one of the preceding claims in which a time averaging filter with a selectable time constant is used to gain knowledge of relatively slow changes in time of the impedance measured
10 spectrum.

23. The method according to claim 8, or any one of claims 9 to 22 each as appended to claim 8, in which provision is made to record the compensation transfer function and its change in time.

24. The method according to any one of the preceding claims in which
15 time averaging of the measured frequency response is used to reduce the measurement noise.

25. A method according to claim 12, or any one of claims 13 to 24 each as appended to claim 12, in which the electrodes are integrated in a micro fluidic or micro analytical chip, the electrodes being either in
20 contact or not in contact with the fluid.

26. A method according to any one of the preceding claims in which the samples are arranged in an array.

27. A method according to any one of the preceding claims in which a change in the system measured impedance spectrum occurs and is recorded.
28. A method according to any one of the preceding claims in which a
5 transient relevant change in the sample measured impedance spectrum occurs during less than 100 millisecond.
29. A method according to claim 12 or any one of claims 13 to 28 each as appended to claim 12 in which at least one electrode has one or more dimensions smaller than 100 micrometers.
- 10 30. A method according to any one of the preceding claims in which the sample under test is a sample comprising a suspension of one or more particles or biological cells, micro-organisms, viruses, molecules, vesicles, or any suspension of organic material.
31. A method according to claim 30 in which cells are passing in a
15 small channel.
32. A method according to claim 30 in which the sample is one or more cells over or on a surface.
33. A method according to claim 31 in which the sample is one or more cells over one or more holes.
- 20 34. A method according to claim 30 in which the sample is one or more cells measured in a patch clamp.

35. A method according to claim 27 in which applying a chemical or biological reactant (gaseous, solid, liquid) triggers or results directly or indirectly in an impedance change of the system.
36. A method according to any one of the preceding claims in which the
5 sample comprises a liquid in a different liquid, a gas in a liquid, an emulsion, particles in a liquid, molecules in a liquid, contaminant in a liquid, or protein in a liquid.
37. A method according to any one of claims 1 to 35 in which the
10 sample comprises a fuel, oil, an organic or inorganic liquid, a biological liquid such as blood or milk, diluted or not, or water, which may or may not contain contaminants.
38. A method as claimed in any one of claims 12 to 15 in which measurement of the contaminant is based on the measured response.
39. A method according to any one of claims 12 to 15 in which the
15 sample is a biological tissue, in vitro or in vivo, measured by electrical impedance tomography.
40. A method according to any one of the preceding claims and used to measure gas level or glucose level in blood or other biological fluid.
41. A method according to any one of claims 12 to 15 in which the role
20 of the electrodes are changed in time.
42. A method according to any one of claims 1 to 24 in which the sample is investigated for corrosion, defects or cracks or material defects.

43. A method according to any one of claims 1 to 24 in which the sample is a polymer based sensor in a gas or liquid.
44. A method according to any one of claims 1 to 24 in which the sample comprises a bilayer lipid membrane.
- 5 45. A method according to any one of claims 1 to 24 in which the sample comprises a monolayer of biological material, such as lipid, protein, fatty acid, organic molecule, on an electrode.
46. A method according to claim 45 in which the monolayer is a self assembled monolayer.
- 10 47. Apparatus for measuring, by the method according to any one of the preceding claims, the AC electrical parameters of a sample, as hereinbefore defined, the apparatus comprising a generator for generating a pseudo random binary sequence, a digital to analogue (D/A) unit or equivalent, or other means to output the sequence in digital or analogue
- 15 format, first connection means for connecting the output of the generator to the input of the D/A, sample input connection means adapted to apply the output of the D/A to the sample, sample output connection means for connecting a sample output to an analogue to digital converter (A/D) or equivalent, and an output processing unit being configured for subjecting
- 20 the output of the A/D to a Hadamard Transform followed by a Fast Fourier Transform (FFT) for producing in use a measurement of the complex impedance frequency response of the sample.
48. Apparatus as claimed in claim 47 comprising a compensation transfer function unit capable, when configured, of modifying the output
- 25 of the generator, the compensation transfer function unit being connected or connectable between the output of the generator and the D/A unit.

49. Apparatus as claimed in claim 47 comprising a compensation transfer function unit capable, when configured, of modifying the output of the output processing unit, the compensation function unit being connected or connectable to the output of the output processing unit.

5 50. An apparatus according to any one of claims 47 to 49 in which one or more sequence generators, and/or one or more digital-to-analogue converters and/or one or more analogue-to-digital converters are integrated *in* an integrated circuit.

10 51. An apparatus according to claim 49 as appended to claim 48 or 49 in which one or more sequence generators, one or more compensation transfer function units, and/or one or more digital-to-analogue converters and/or one or more analogue-to-digital converters are integrated in an integrated circuit.

15 52. An apparatus according to claim 50 or claim 51 in which the measured system response is additionally processed in use by the said integrated circuit.

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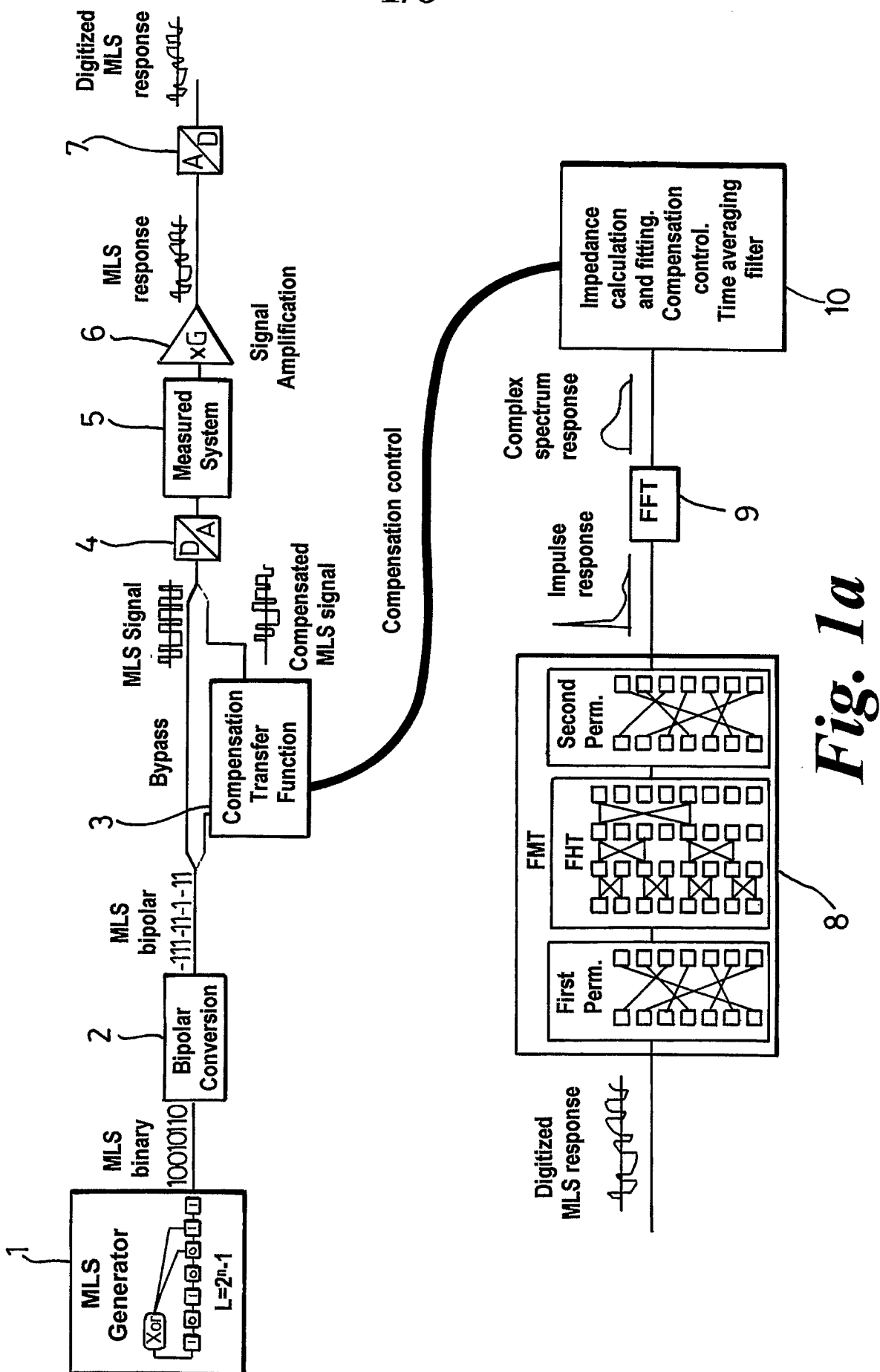


Fig. 1a

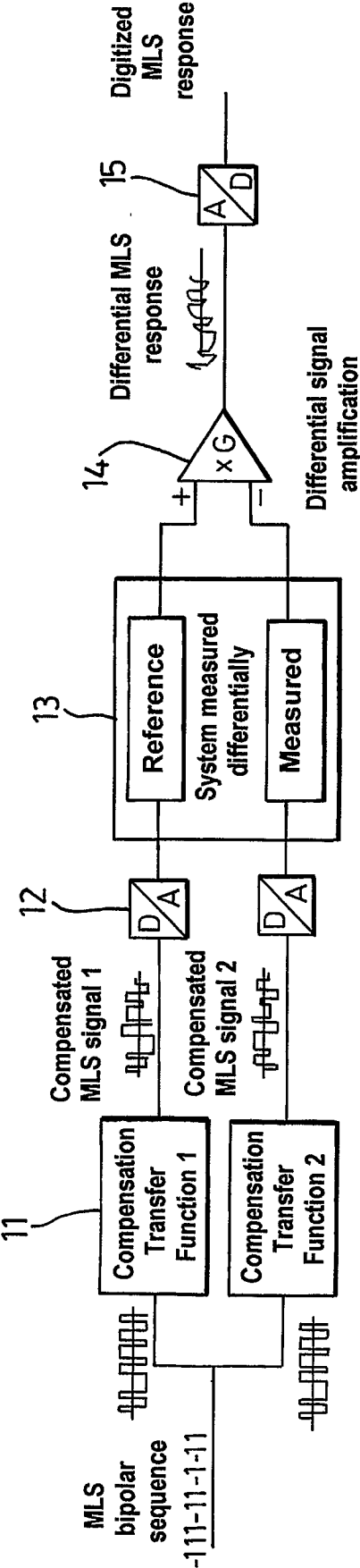
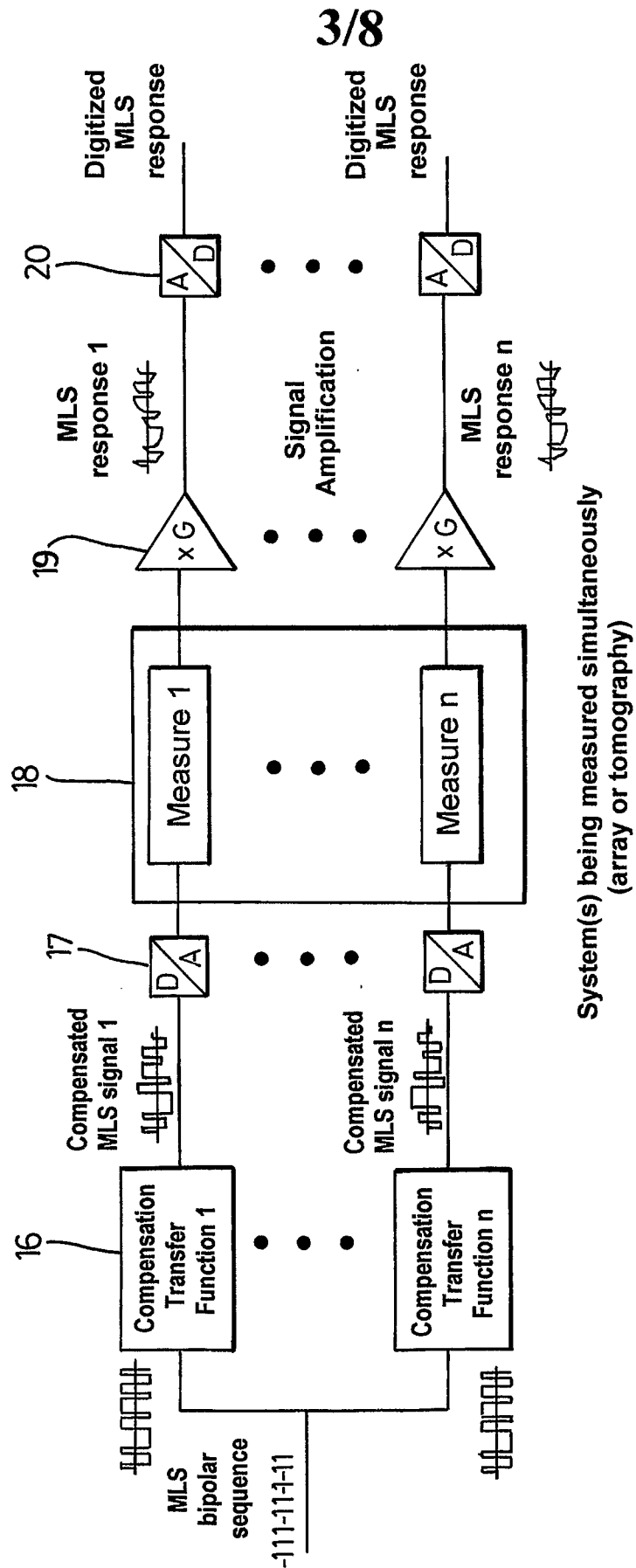


Fig. 2

**Fig. 3**

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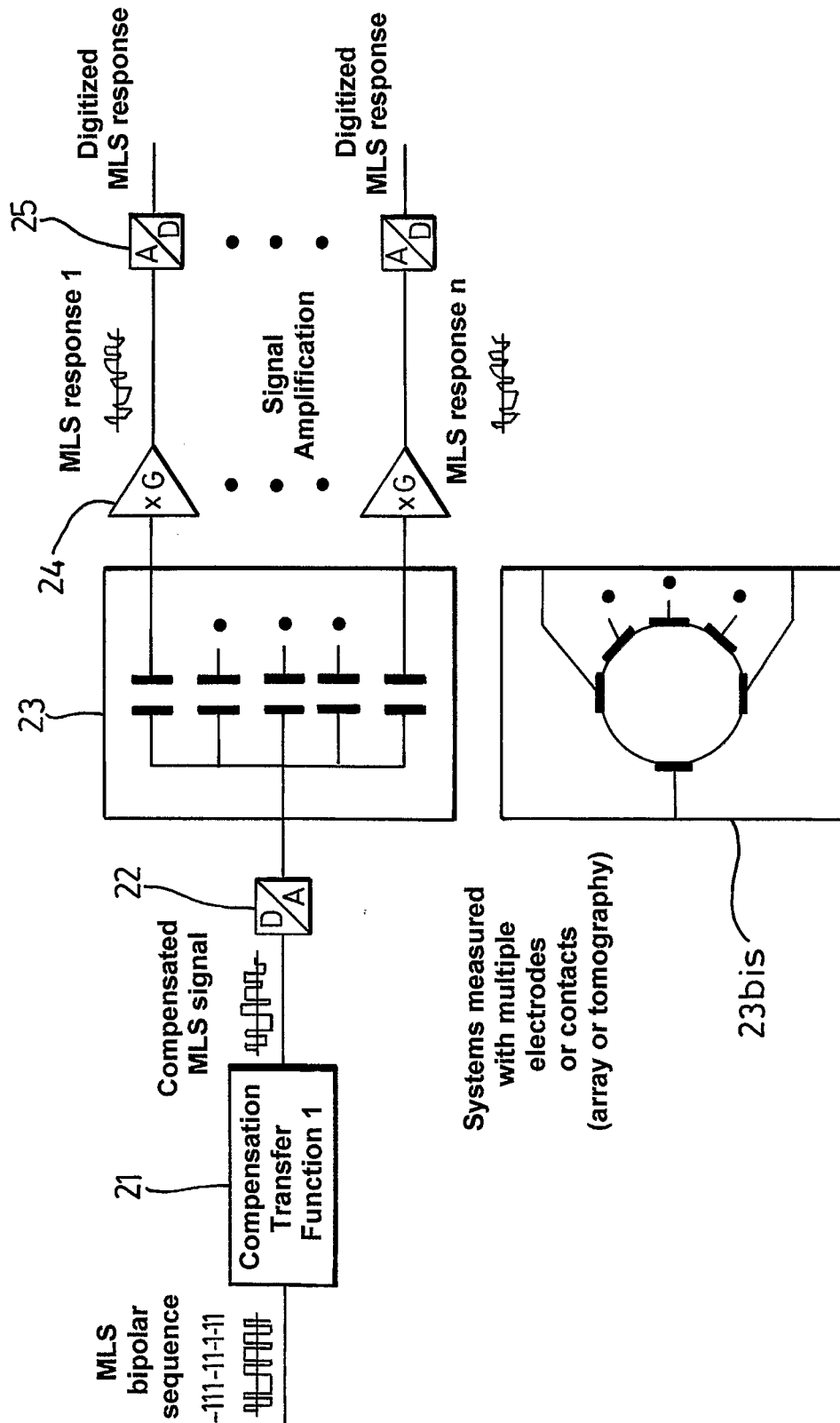
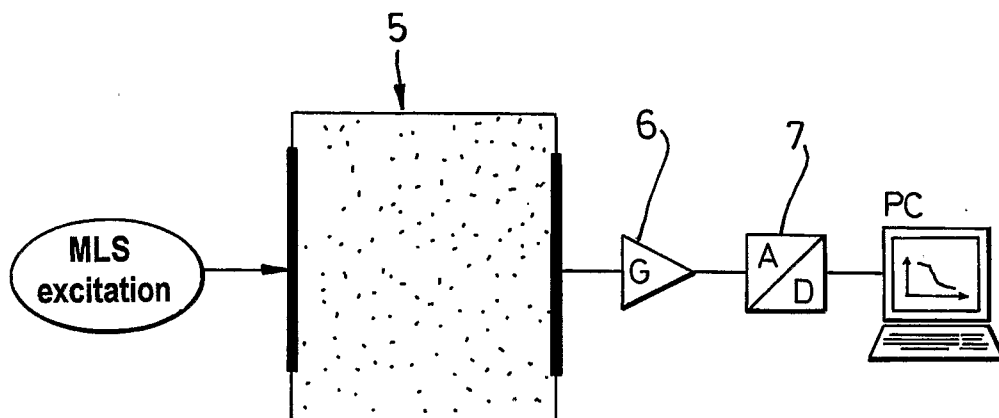
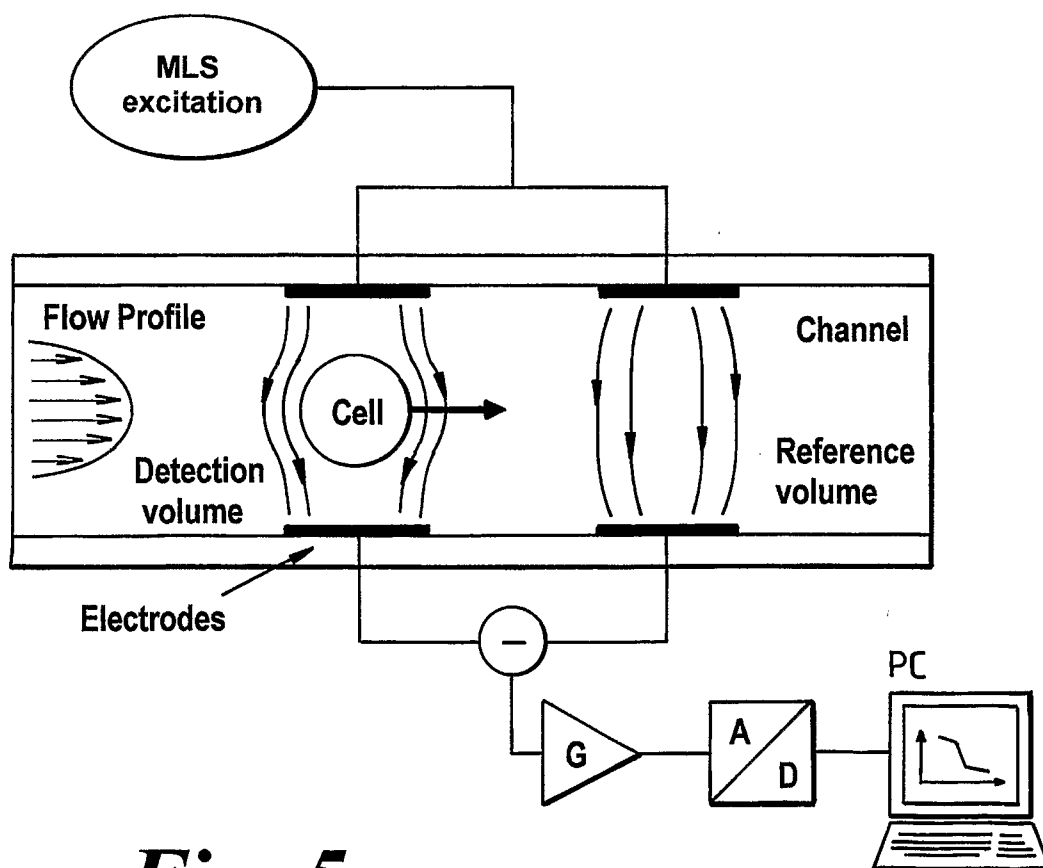
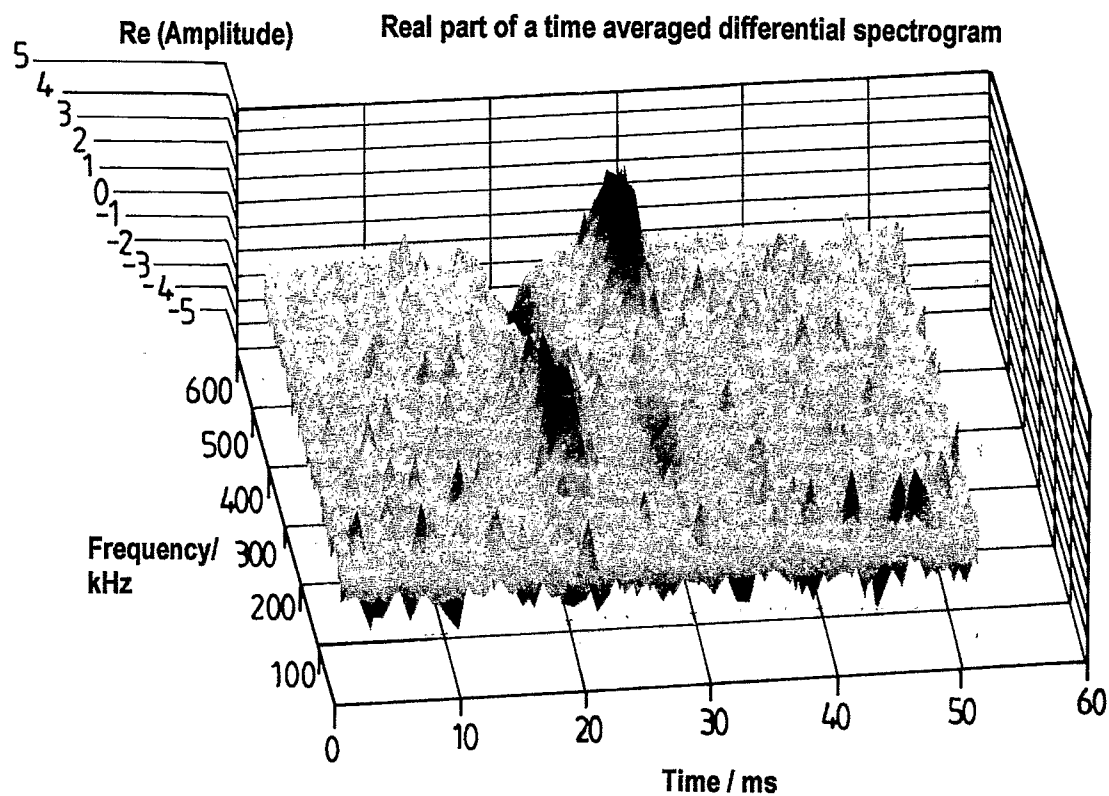
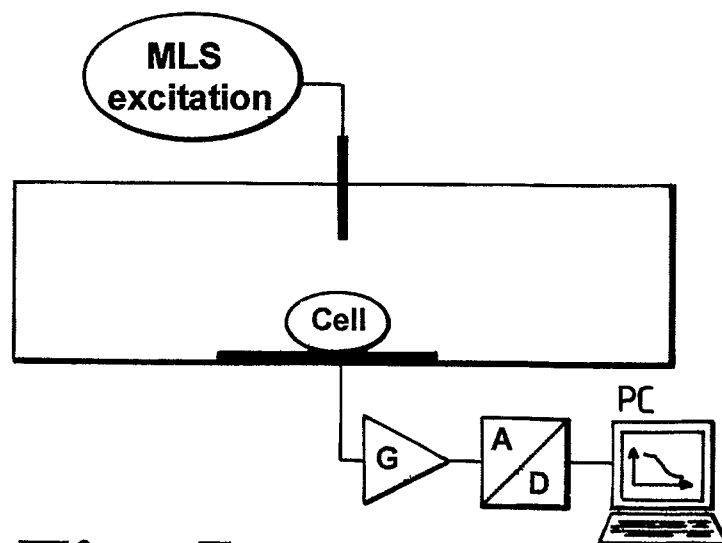


Fig. 4

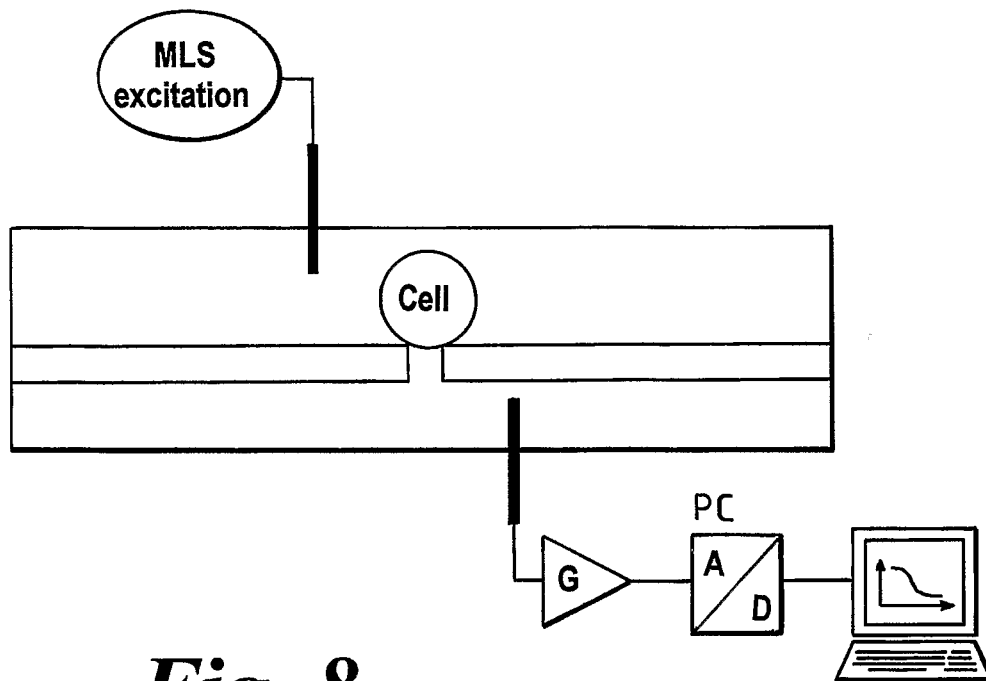
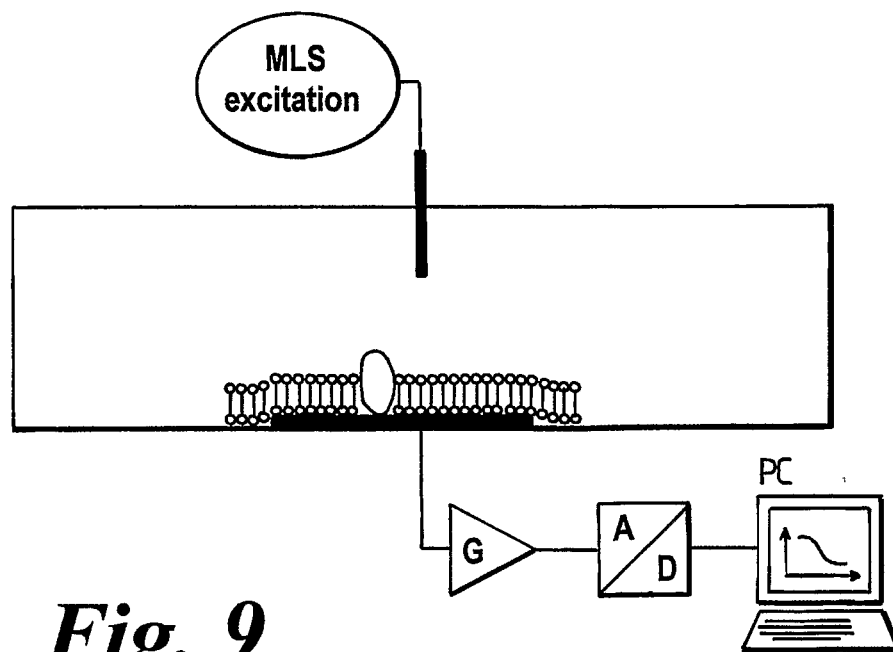
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*Fig. 1b**Fig. 5*

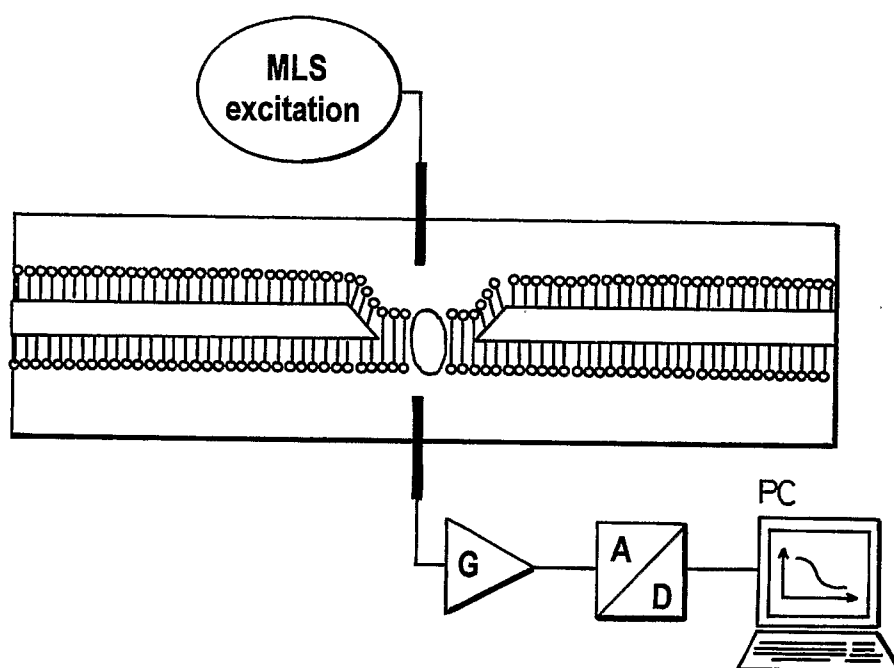
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*Fig. 6**Fig. 7*

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*Fig. 8**Fig. 9*

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*Fig. 10*

INTERNATIONAL SEARCH REPORT

International application No
PCT/GB2006/004185

A. CLASSIFICATION OF SUBJECT MATTER
INV. G01R27/28

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

GOIR

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, WPI Data, INSPEC, COMPENDEX, IBM-TDB

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document with indication, where appropriate, of the relevant passages	Relevant to claim No
Y	WO 97/18467 A (AROMASCAN PLC [GB]; AMRANI MOHAMMED EL HASSAN [GB]; PAYNE PETER ALFRED) 22 May 1997 (1997-05-22) cited in the application abstract; figures 1,5,6 page 1, line 1 - page 2, line 27 page 3, line 26 - line 27 page 7, line 13 - line 18 page 9, line 8 - line 20; claims 1,9 ----- -/-	1-52

☒ Further documents are listed in the continuation of Box C

☒ See patent family annex

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"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

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Date of the actual completion of the international search

14 March 2007

Date of mailing of the International search report

21/03/2007

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Binger, Bernard

INTERNATIONAL SEARCH REPORT

International application No
PCT/GB2006/004185

(^Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	BORISH J ET AL: "AN EFFICIENT ALGORITHM FOR MEASURING THE IMPULSE RESPONSE USING PSEUDORANDOM NOISE" JOURNAL OF THE AUDIO ENGINEERING SOCIETY, AUDIO ENGINEERING SOCIETY, NEW YORK, NY, US, vol. 31, no. 7, July 1983 (1983-07), pages 478-488, XP009060089 ISSN: 0004-7554 paragraphs [0000], [0001], [0005], [06.1], [06.4], [06.5], [0008] -----	1-52

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Information on patent family members

International application No

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