Title: APPARATUS AND METHODS FOR ANALYSING BODILY TISSUE

Abstract: There is disclosed an apparatus for analysing bodily tissue comprising: means (10) for making clinically relevant measurements of said bodily tissue using at least two analysis techniques selected from the group comprising: photoacoustic analysis; fluorescence spectroscopy; Raman spectroscopy; near infra-red spectroscopy; optical confocal microscopy; and optical chromaticity; in which said means (10) for making clinically relevant measurements comprises a plurality of light sources (12, 14) which provide electromagnetic radiation used to perform the analysis techniques; and further comprising a light guiding system (16, 18) coupled to each of the light sources (12, 14) which channels electromagnetic radiation from each of the light sources to the bodily tissue under analysis substantially along a common pathway so that electromagnetic radiation from the light sources (12, 14) emerges from the light guiding system (16, 18) at a common point.
— before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments

For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.
Apparatus and Methods For Analysing Bodily Tissue

This invention relates to apparatus and methods for analysing bodily tissue.

There are a large number of analytical techniques which have been utilised, with varying degrees of success, for in vivo clinical monitoring. For example, there is considerable interest in medical applications of ultrasound and photoacoustic waves. This is because, in contrast to interrogating techniques such as X-ray imaging and scintigraphy, ultrasound offers the possibility of interrogating a subject with no harmful side effects. Ultrasound imaging of cardiac structures, the vascular systems, the foetus and uterus, abdominal organs such as the liver, kidneys and gall bladder is known. Additionally, ultrasound measurements of the eye are known. It should be noted that, in addition to applications which are concerned solely with the generation of images, ultrasound techniques can provide information, albeit limited information, about the nature of the tissue being interrogated. Principally, such information relates to the elasticity of the interrogated tissue. However, it is not possible to provide detailed information concerning the state of the tissue under interrogation.

Henceforth the term “photoacoustic analysis” will be used, and it is understood that this term refers to analyses which employ the detection of optically generated ultrasound (also known as photoacoustic waves).

Another analytical technique of clinical relevance is Fourier Transform near infra-red (FT-NIR) spectroscopy. Two spectral bands of clinical relevance are the 700-1000nm region and the 1000-5000nm region. The 700-1000nm region is used to measure changes in oxyhaemoglobin, deoxyhaemoglobin and cytochrome oxidase, yielding information on haemoglobin, tissue deoxygenation and changes in blood flow. The 1000-
5000nm region facilitates measurement of other physiological parameters, such as glucose, birubin and biliverdin, that are important in many clinical situations such as diabetes, liver failure and intensive care. The use of FT-NIR in practical circumstances has been facilitated by the development of a compact spectrometer which can be used for real time clinical *in vivo* monitoring purposes. However, the information provided by such devices is limited by the fact that these clinical devices only utilise four to six discrete wavelengths for analysis.

Another analytical technique of clinical relevance is fluorescence microscopy, which is a well-established tissue spatial imaging technique. Differences in endogenous, *in vivo*, fluorescence of healthy and diseased tissue, within the same patient, also afford a highly sensitive, non-intrusive, way of differentiating when only healthy tissue is present. As an alternative, added fluorescers, which target unhealthy tissue, can be used to differentiate unhealthy tissue. This is particularly useful when combined photodynamic therapy and surgery are the preferred option for treatment and when, for example, fluorescent photosensitisers are being used.

The present inventors have recognised that there is a long felt need for a clinical technique which can reliably perform a tissue identification function. In particular, it would be desirable to be able to identify, with good spatial resolution, healthy tissue from diseased tissue. A non-limiting example of an application in which such a function would be desirable is surgery. When surgical techniques are used to remove diseased tissue, it is highly desirable to remove only the diseased tissue, and not any surrounding, healthy tissue. Typically, however, if the surgeon has any doubt about the nature of the tissue, the surgeon takes a cautious approach, and removes an excess of tissue, inclusive of healthy tissue. It would be highly desirable to be able to differentiate healthy tissue from diseased tissue with good spatial resolution during surgical procedure. This is particularly important for techniques such as brain surgery, where the tissue in question
performs a vital and irreplaceable function. However, none of the techniques discussed above are wholly satisfactory for performing such tissue identification or tissue differentiation functions.

Furthermore, it is highly desirable that any technique intended for use in a clinical environment is compact, reliable, convenient, easy to operate and that the results provided by the technique are readily comprehensible to an operator who is not skilled in analytical science.

The present invention addresses the above mentioned needs, desires and problems.

According to a first aspect of the invention there is provided apparatus for analysing bodily tissue comprising:

means for making clinically relevant measurements of said bodily tissue using at least two analysis techniques selected from the group comprising: photoacoustic analysis; fluorescence spectroscopy; Raman spectroscopy; near infra-red spectroscopy; optical confocal microscopy; and optical chromaticity;

in which said means for making clinically relevant measurements comprises a plurality of light sources which provide electromagnetic radiation used to perform the analysis techniques;

and further comprising a light guiding system coupled to each of the light sources which channels electromagnetic radiation from each of the light sources to the bodily tissue under analysis substantially along a common pathway so that electromagnetic radiation from the light sources emerges from the light guiding system at a common point.
By combining the above described techniques, tissue differentiation or even identification is possible. Furthermore, the apparatus is practical and convenient to use because the laser or light sources which initiate each of the analysis techniques are coupled to the tissue under investigation by a common light guiding system. Conveniently, the light guiding system may be held in a single hand of an operator, who can thereby control the position of the light guiding system with respect to the tissue and thus control which portion of tissue is interrogated. This leaves the other hand of the operator free for performing other tasks, for example surgical techniques. Any combination of the above described is within the scope of the invention.

The light guiding system may comprise optical fibre, which might be multi-mode optical fibre.

Alternatively, the light guiding system may comprise a waveguide.

Preferably, the light sources comprise at least one laser source, which might be, for example, a Nd-YAG, Ar-ion or diode laser. The precise selection of laser source is dependent on the precise analytical techniques selected. One of the techniques may comprise laser induced fluorescence. Alternatively, or additionally, other light sources, such as krypton arc lamps, xenon arc lamps and light emitting diodes (LEDs), might be used.

In embodiments in which the means for making clinically relevant measurements uses photoacoustic analysis, said means may comprise a probe head having ultrasonic transducer means disposed on the light guiding system in the region in which the electromagnetic radiation emerges from same.
The means for making clinically relevant measurements may use Fourier Transform near infra-red spectroscopy.

In embodiments in which fluorescence spectroscopy is used, the means for making clinically relevant measurements may use fluorescence microscopy.

The apparatus may comprise analysis means for analysing the measurements of said bodily tissue. The analysis means may comprise artificial intelligence means, which may comprise a neural network. An advantage with such artificial intelligence techniques is that they can be trained to robustly recognise certain tissue types, e.g., diseased tissue, without any skilled intervention from the operator.

The analysis means may be adapted to differentiate between measurements corresponding to healthy tissue and measurements corresponding to diseased tissue. The analysis means may perform subtraction of fluorescent images of healthy tissue from fluorescent images of diseased tissue. The apparatus may comprise means for displaying the results of the subtraction, preferably a visual display such as a screen or monitor.

The apparatus may further comprise a therapeutic laser source which provides electromagnetic radiation having characteristics suitable for therapy of the bodily tissue under analysis, which radiation is channelled by the light guiding system to said bodily tissue.

According to a second aspect of the invention there is provided a method for analysing bodily tissue comprising the steps of:

channelling electromagnetic radiation from a plurality of light sources to the bodily tissue under analysis substantially along a common pathway using a
light guiding system so that said electromagnetic radiation emerges from the light guiding system at a common point; and

making clinically relevant measurements of said bodily tissue with said electromagnetic radiation using at least two analysis techniques selected from the group comprising: photoacoustic analysis; fluorescence spectroscopy; Raman spectroscopy; near infra-red spectroscopy; optical confocal microscopy; and optical chromaticity.

The apparatus of the first aspect of the invention may be used in conjunction with the method of the second aspect of the invention.

Methods and apparatus in accordance with the invention will now be described with reference to the accompanying drawings, in which:-

Figure 1 is a schematic diagram of apparatus according to the invention; and

Figure 2 is a perspective view of optical fibre and a photo acoustic probe unit.

Figure 1 shows a non-limiting embodiment of apparatus of the present invention. The apparatus, shown generally at 10, comprises one or more pulsed laser sources 12. Optionally, a continuous light source 14, such as a CW laser source or an arc lamp, may be employed. Light L1 produced by the laser sources 12 is directed through a beam splitter 16 and into an optical fibre 18. Typically, the optical fibre 18 is a multimode fibre. From the optical fibre 18, light L1 is delivered to a sample surface 20 where medical
diagnosis is required. The light L1 irradiates the sample 20, from which return signals of an optical or other electromagnetic nature, or ultrasonic nature are monitored.

Optical signals, L2, emanating from the sample 20 are transmitted back along the same optical fibre 18, and are then transmitted via the beamsplitter 16 to a combination of optical instruments 22, 24, 26. A range of techniques, such as FT-NIR (Fourier transform near infrared) spectroscopy, fluorescence microscopy, and optical confocal microscopy may be used. Thus, in this embodiment, the instruments comprise a FT-NIR diagnostic system 22, a spectrograph 24 for fluorescence microscopy and an optical confocal microscopy system 26.

Photoacoustic signals may be detected by transduction using a photoacoustic probe unit 28. The electrical signals generated thereby are transmitted to a photoacoustic diagnostic system 30. The signals may relate to a forward viewing probe, a sideways viewing probe, or both. Any combination of these systems may be used.

Sources of light, L1, also may include light sources that are used at the same time for therapeutic treatment.

Figure 2 shows further detail of the optical fibre cable 18 and probe unit 28. The cable 12 is typically ca. 1.5m long, although this length is in no way limiting.

Laser radiation is introduced into the optical fibre 18, which may of ca. 600μm OD, via a connector 32. The optical fibre 18 passes through a signal manifold 34 which comprises a connection 34a which is in electrical connection to the transducer means 36. A cable 38 runs between the manifold 34 and a housing 40, the cable 38 containing the optical fibre 18 and electrical signal leads from the housing 40. Light L1 from the sources 12, 14 emerges at a common source point 18a, which source point is one
end of the optical fibre 18. The housing 40 comprises a perspex shell, in which the ultrasonic transducer means 36 is located. Although the ultrasonic transducer means 36 might comprise a piezoelectric ceramic, it is preferred to use a piezoelectric polymeric material, preferably polyvinylidene fluoride (PVDF) or, most preferably, its copolymer with trifluoroethylene (TrFE). The PVDF is a film (Pennwalt Kynar film type 5028 NAD), the rear face of which is located on a conductive silver loaded epoxy support (Circuit Works RTM, available from RS). The use of silver loaded epoxy is preferred because i) it exhibits advantageous mechanical properties of stiffness, acoustic impedance and good adhesion to PVDF and ii) it exhibits excellent electrical conductivity compared to conventional epoxy materials, resulting in increased detection sensivity. A wire makes electrical contact with the epoxy support, the main body of the wire being located in cable 38. The housing 40 is coated with a silver loaded paint. The optical fibre 18 passes through the housing 40, preferably coaxially therewith.

The electrical signal obtained from transduction of the photoacoustic signal is passed to the photoacoustic diagnostic system 30 via connection 34a.

This system may perform simple A-scans, or produce B- or C-scans for imaging and diagnosis. Linked to analysis means such as a computer system 42, neural networks may be used to help diagnosis. The same computer system 42 may then be used to combine measurements from the optical diagnosis systems to help enhance the diagnosis. In fact, the “data fusion” which can be achieved by using a plurality of analysis techniques is a powerful aspect of the invention. It has been found that the analysis techniques utilised in the present invention have complementary facets which, in combination, provide much improved analysis of bodily tissue. For example, photoacoustic investigations provide information about elasticity, and infra-red techniques provide some depth penetration.
Artificial intelligence such as a neural network can be taught to recognise the difference between healthy and diseased tissue. It may be possible to provide a simple two level system i.e., a GO/NO GO system, which identifies tissue either as healthy or diseased. Alternatively, more detailed information might be extracted.

The analysis means may use computer subtraction of fluorescent images from a patient on a screen for a surgeon to assess (i) a region of interest to provide healthy tissue image, (ii) a region of interest to provide an unhealthy tissue image (iii) a visible and computer comparison differentiating unhealthy tissue from healthy tissue.

Examples of tissue which might be analysed include brain, liver, kidney and muscle tissue.

It is possible to utilise further analysis techniques in combination with the above described techniques. For example, NMR, ESR and mass spectrometry might be contemplated. It may be possible to integrate the present invention with existing medical and surgical techniques, such as robotic surgery.
CLAIMS

1. Apparatus for analysing bodily tissue comprising:

5 means for making clinically relevant measurements of said bodily tissue using at least two analysis techniques selected from the group comprising: photoacoustic analysis; fluorescence spectroscopy; Raman spectroscopy; near infra-red spectroscopy; optical confocal microscopy; and optical chromaticity;

10 in which said means for making clinically relevant measurements comprises a plurality of light sources which provide electromagnetic radiation used to perform the analysis techniques;

and further comprising a light guiding system coupled to each of the light sources which channels electromagnetic radiation from each of the light sources to the bodily tissue under analysis substantially along a common pathway so that electromagnetic radiation from the light sources emerges from the light guiding system at a common point.

2. Apparatus according to claim 1 in which the light guiding system comprises optical fibre.

3. Apparatus according to claim 2 in which the light guiding system comprises multi-mode optical fibre.

25 4. Apparatus according to claim 1 in which the light guiding system comprises a waveguide.
5. Apparatus according to any of claims 1 to 4 in which the light sources comprise at least one laser source.

6. Apparatus according to claim 5 in which one of the analysis techniques selected comprises laser induced fluorescence spectroscopy.

7. Apparatus according to any of claims 1 to 6 in which the means for making clinically relevant measurements uses photoacoustic analysis, and comprises a probe head having ultrasonic transducer means disposed on the light guiding system in the region in which the electromagnetic radiation emerges from same.

8. Apparatus according to any previous claim in which the means for making clinically relevant measurements uses Fourier Transform near infra-red spectroscopy.

9. Apparatus according to any previous claim in which the means for making clinically relevant measurements uses fluorescence microscopy.

10. Apparatus according to any previous claim comprising analysis means for analysing the measurements of said bodily tissue.

11. Apparatus according to claim 10 in which the analysis means comprise artificial intelligence means.

12. Apparatus according to claim 11 in which the artificial intelligence means comprises a neural network.
13. Apparatus according to any of claims 10 to 12 in which the analysis means is adapted to differentiate between measurements corresponding to healthy tissue and measurements corresponding to diseased tissue.

14. Apparatus according to claim 13 in which the analysis means performs subtraction of fluorescent images of healthy tissue from fluorescent images of diseased tissue.

15. Apparatus according to claim 14 comprising display means for displaying the results of the subtraction.

16. Apparatus according to any previous claim further comprising a therapeutic laser source which provides electromagnetic radiation having characteristics suitable for therapy of the bodily tissue under analysis, which radiation is channelled by the light guiding system to said bodily tissue.

17. A method for analysing bodily tissue comprising the steps of:

  channelling electromagnetic radiation from a plurality of light sources to the bodily tissue under analysis substantially along a common pathway using a light guiding system so that said electromagnetic radiation emerges from the light guiding system at a common point; and

  making clinically relevant measurements of said bodily tissue with said electromagnetic radiation using at least two analysis techniques selected from the group comprising: photoacoustic analysis; fluorescence spectroscopy; Raman spectroscopy; near infra-red spectroscopy; optical confocal microscopy; and optical chromaticity.
### INTERNATIONAL SEARCH REPORT

#### A. CLASSIFICATION OF SUBJECT MATTER

**IPC 7** A61B5/00

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)

**IPC 7** A61B

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

#### C. DOCUMENTS CONSIDERED TO BE RELEVANT

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<td>X</td>
<td>WO 99 27848 A (ABBOTT LAB) 10 June 1999 (1999-06-10) abstract</td>
<td>1-16</td>
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<td>page 5, line 22 - line 23 page 10, line 30 - page 11, line 8 page 18, line 14 - page 19, line 19 page 30, line 28 - line 34 page 37, line 23 - page 38, line 3 page 45, line 3 - line 6 figure 7</td>
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<td>X</td>
<td>US 5 293 872 A (LIU CHENG H ET AL) 15 March 1994 (1994-03-15) column 8, line 14 - column 9, line 28 figures 9,10</td>
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* Special categories of cited documents:

**A** document defining the general state of the art which is not considered to be of particular relevance

**E** earlier document but published on or after the international filing date

**L** document which, although not referring to the priority claimed, is essential for the understanding of the subject matter

**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

**X** 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

**X** document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

**X** document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

**X** document member of the same patent family

#### Date of the actual completion of the International search

11 December 2002

#### Date of mailing of the International search report

20/12/2002

#### Name and mailing address of the ISA

European Patent Office, P.B. 5616 Patentlaan 2 NL - 2280 HV Rijswijk, Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax (+31-70) 340-3016

Authorized officer

Lohmann, S

Form PCT/GB01 (second sheet) (July 1999)
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<td>US 5 348 002 A (CARO RICHARD G) 20 September 1994 (1994-09-20) column 7, line 30 - line 47 column 11, line 4 - line 15 column 19, line 29 - line 35 figures 1-3</td>
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INTERNATIONAL SEARCH REPORT

Box I  Observations where certain claims were found unsearchable (Continuation of Item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1.  
   
   [X] Claims Nos.: 17
   
   because they relate to subject matter not required to be searched by this Authority, namely:

   Rule 39.1(iv) PCT - Diagnostic method practised on the human or animal body

2.  

   [ ] Claims Nos.:  
   
   because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:

3.  

   [ ] Claims Nos.:  
   
   because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II  Observations where unity of invention is lacking (Continuation of Item 2 of first sheet)

This International Searching Authority found multiple inventions in this International application, as follows:

1.  

   [ ] As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2.  

   [ ] As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3.  

   [ ] As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4.  

   [ ] No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.
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