

Fig. 2. Schematic diagram of the electronic amplifier.

with the movement of the lever is heard. This will indicate that at that precise moment the sensitivity of the amplifier has reached the same value as the previously chosen light intensity. With the sensitivity knob set at this point the amplifier is calibrated to that particular value and will not operate with lower intensities of light.

The sensitivity range of the amplifier runs from a maximum of 0.02 lux to a minimum of 160 lux. At its minimum sensitivity the amplifier will work only with light intensities of 160 lux or over. At its maximum even light intensities of 0.02 lux will make the apparatus work. Once the amplifier has been calibrated to the desired light intensity the sample is inserted in front of the light source. If the interposition of the sample makes the intensity of the light reaching the photocell of the amplifier drop below the sensitivity level for which the amplifier was calibrated, the up and down movement of the lever does not produce any change in sound. The intensity of light has to be slowly increased till the movement of the lever in front of the light source will produce the peep-like sound again. This indicates that, at that particular moment, an intensity of light equal to that for which the amplifier was calibrated has reached the photocell of the amplifier. The intensity of light is fixed at this particular value. The sample is then taken off and the optic probe is disconnected from the photocell of the amplifier and connected to the photometer (the other end of the optic probe still facing the light source) to read the new increased intensity of light which was able to make the system work again. From these 2 values the percent light transmitted can be calculated according to the formula  $I_2:I_1\times 100$ ; where the first intensity is represented by the values with which the amplifier was calibrated and the second intensity by the values the light reached at the start of the change in sound after the interposition of the sample. In order to evaluate the validity of the method, measurements for light transmission were taken using both the usual method, in which the intensity of the light remains costant, and this audio method in which the intensity of light has to be increased instead, so that the same amount

of light can reach the photocell after the interposition of the sample. Kodak neutral density filters were used as standards. The results, as percent light transmission, are shown in the table, A 5 V, 0.8 A bulb, powered by a battery and connected to a rheostat, was used as a light source. In using the usual method the free end of the optic probe was permanently connected to the photometer and the 2 different intensities of light with and without the interposition of the filters were obtained, while the intensity of light remained constant throughout the measurements.

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## Light transmission. II. Instruments for making in-vivo light transmission measurements

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Summary. 2 instruments for making in vivo light transmission measurements are described. A special device makes it possible to measure the thickness of the tissue as well.

The increased therapeutic use of light<sup>3-6</sup> and the awareness of its potential deleterious effects have given a new interest to the study of its power to penetrate biological tissues. The

various methods used to measure light transmission<sup>7,8</sup> do not offer the possibility of making in-vivo measurements. The purpose of this study is to present 2 instruments for making light transmission measurements through living tissues

The 2 instruments, made of plexiglas, use fibreoptic probes to conduct light. A high intensity fan-cooled illuminator is employed as light source. Light intensities are read on a photometer. The instruments have the tips of the 2 fibreoptic probes facing each other at right angles, so that the beam of light can impinge on the bevelled tip of the probe going to the photometer.

The instrument shown in figure 1 has the 2 tips at a fixed distance. Other instruments of the same type differ in the height of the cone holding the probe going to the photometer and in the distance between the 2 tips. The instrument uses 2 fibreoptic probes each having an internal diameter of 4.5 mm and a length of 122 cm. The longer horizontal hole houses the probe coming from the illuminator. A slot is provided to accomodate the filter. The smaller horizontal hole below may be used as an alternative for the axis holding a rotating filter holder. The cone-shaped vertical part accomodates the probe going to the photometer. Its metal end is tight-fitted, bevelled and faces directly the tip of the other probe coming from the illuminator.

The prototype shown in figure 2 makes use of a small bundle of glass fibreoptics which at one end is inserted into a syringe needle and at the other end is provided with a ferrule to be connected to the photometer. The receiving end of the fibreoptic bundle is bevelled and shaped like the tip of the syringe needle in which it is fed. It is suitable for insertion under the skin or inside soft tissues. The diameter and length of the syringe needle to be used depend on the tissue to be measured.

The instrument is equipped also to take measurements of the thickness of the tissue or the depth reached by the inserted needle. It is made up of a 2.5 \times 2.5 cm supporting bar, 9 cm long, with a hole throughout its length the diameter of which varies according to the source of light it has to accomodate, which may be either an optic probe connected to an illuminator or a light bulb connected to a battery. An L-shaped metal stick, 2.5 cm in width, slotted in the center, is fastened with 2 hand screws alongside the supporting bar. Its shorter arm holds a universal joint device provided with a small rectangular plexiglas block in which the specially prepared syringe needle is encased. The slots in both arms of the L-shaped metal stick make both up and down and sideways movements possible, so that the tip of the optic probe going to the photometer may be adjusted accordingly. Finer adjustments are obtained by the rotating action of the universal joint so that the beam of light can be made to impinge exactly on the bevelled tip of the needle. A plexiglas stick 2.5 cm wide and 14 cm long,

slotted in the center, is secured below the supporting bar by means of a screw. 2 pins attached beneath the supporting bar and going through the slot act as guides providing, together with the screw when is not tightened, a straight

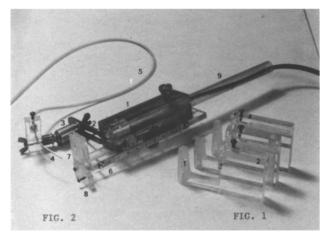


Fig. 1. 1: Holder for optic probe going to photometer. 2: Holder for optic probe coming from illuminator. 3: Slot for filter.

Fig. 2. 1: Supporting bar. 2: L-shaped metal stick. 3: Universal joint. 4: Syringe needle with optic fibres. 5: Optic probe going to photometer. 6: Sliding-graded plexiglas stick with filter holder. 7: Slot for filter. 8: 'Center finder' device. 9: Plastic tube housing the light bulb.

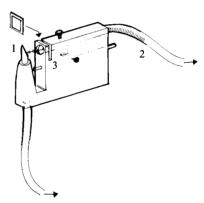


Fig. 3. Instrument with fixed distance. 1: Bevelled tip of the optic probe going to the photometer. 2: Optic probe coming from the illuminator. 3: Slot for the filter. 4: Filter.

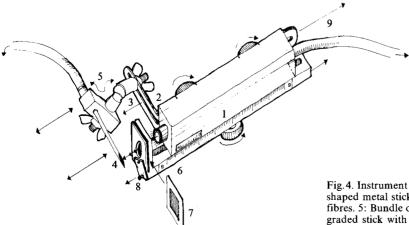


Fig. 4. Instrument with adjustable distance. 1: Supporting bar. 2: L-shaped metal stick. 3: Universal joint. 4: Syringe needle with optic fibres. 5: Bundle of optic fibres going to the photometer. 6: Sliding-graded stick with filter holder. 7: Filter. 8: 'Center finder' device. 9: Optic probe coming from the illuminator.

sliding movement to the stick, which is provided in front with a special holder for the filter.

An 8 cm long graduated metal ruler is attached alongside the sliding stick and just above, aligned with it, at the end of the supporting bar, a vernier is also attached. A 'center finder' in front of the stick makes it possible to take more accurate measurements. It consists of a small rectangular plexiglas plate provided with a small pin-like screw which, when the plate is in the up-position, is aligned with the source of light and faces the bevelled tip of the needle. When light transmission measurements are taken the plate is rotated down so as not to interfere with the beam of light. When the needle is inserted, for the sliding action of the stick, the head of the pin can be made to touch the surface of the tissue and subsequently the bevelled tip of the needle, when the latter is withdrawn. The difference between the 2 distances, read on the graded ruler with the help of the vernier, represents the thickness of the tissue or the depth reached by the tip of the needle.

Measurements of light transmission are taken by inserting the bevelled tip of the optic probe into the tissue or cavity or by holding the tissue so that it lies between the tip of the optic probe and the source of light. A first intensity is read with the light impinging directly on the tip of the optic probe going to the photometer. A second intensity is then read with the interposition of the tissue. From these 2 values the percentage light transmission is calculated as previously reported.

The acoustic method described in the first part of this study can also be used with these 2 instruments, provided a rheostat is connected with the power supply in order to regulate the intensity of light. The procedure would be basically the same. The variations in light intensities, to elicit the peep-like sound, would be brought about by rhythmically intercepting the beam of light.

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## A demonstration of the resolution of NMR imaging in biological systems<sup>1</sup>

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Summary. A proton NMR imaging study of several fruit specimens demonstrates the integrity and resolution of this new imaging method.

Macroscopic thin-section images of heteregeneous systems can be produced by modification of the NMR technique<sup>4-6</sup>. The application of NMR imaging methods to the investigation of living intact organisms is of potential value since it is non-invasive, has no known hazard<sup>7</sup>, and the imaging parameter is a powerful measure of molecular level structure and motion, reflecting also variations in biological tissue age, origin and health<sup>8</sup>. We demonstrate here the spatial resolution, image parameter contrast, and geometric accuracy obtainable by the new 'multiple sensitive point' method<sup>9,10</sup> of NMR image formation by reference to 3 fruit specimens. The fruit specimens possess a higher degree of tissue heterogeneity and geometric complexity than can be imitated by artificial phantoms, thus enabling reasonable extrapolation of the results to complex animal structures.

Experimental. In multiple sensitive point imaging, the NMR spectrometer sensitivity is restricted to a thin 'sensitive line' within the sample by application of 2 orthogonal time-dependent magnetic field gradients<sup>9</sup>. A third, static magnetic field gradient, applied in the direction of the sensitive line, enables the distribution of the NMR signal along the line to be determined after time-averaging and Fourier transformation<sup>5</sup>. Scanning the sensitiveline slowly across the imaging plane generates a complete thin cross-sectional image.

Our experiments employed a home-built NMR spectrometer operating at a 30 MHz proton resonance frequency, and a Varian V-7300 electromagnet with 13 cm pole gap. The sample chamber diameter was 8 cm. Image data were stored on computer cassette tape and displayed as beam

intensity on a storage oscilloscope screen. Regions of high proton NMR signal level in the sample are thus rendered as areas of high intensity on the displayed image.

All images were obtained under similar spectrometer operating conditions and consist of  $128 \times 128$  independent picture points. The cross-sectional shape of the sensitive line, which is determined by the magnetic field gradient strengths, was roughly  $3 \times 0.5$  mm. The spatial resolution in the 3rd direction was also 0.5 mm. Each of the 128 sensitive lines was averaged for 3 sec, and the total imaging time was 380 sec. The proton resonance was excited by a steady-state free precession pulse sequence.

Results and discussion. NMR images of an intact apple, satsuma and plum are shown in figures a, c and d. A subsequent section at the level of the imaging plane of the apple, is pictured in figure b. All images demonstrate accurate structure reproduction with spatial resolution approaching, and in some cases equal to the picture element spacing. Regions such as pith, skin, core, pips and flesh are defined and contrasted by intensity variation within the images. Worthy of particular note are the fine septa of average thickness 0.5 mm which define the flesh segments in the satsuma and the approximately 1 mm thick outline of the seed core of the apple, clearly discernible in these images. Note also the differentiation of the oily surface layer of the satsuma skin which possesses a higher proton NMR signal level than the drier sub-surface pithy layer covering the flesh.

These results illustrate the integrity of NMR imaging as a new non-invasive diagnostic tool whereby high resolution