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PROTON MAGNETIC RESONANCE OF HUMAN GINGIVAL TISSUE

A PRELIMINARY REPORT

by

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When a sample is introduced in a magnetic field there occur absorption lines for electromagnetic radiation. These absorption lines are due to the magnetic properties of atomic nuclei and are called nuclear (in this case, proton) magnetic resonance lines. They occur at unusually long wavelengths (5—500 m), the exact value of the wave-length or frequency being dependent on the isotope (nucleus) under study and on the strength of the magnetic field. The magnetic resonance lines for a given isotope undergo important changes when the nature of the sample is varied. For example, the proton resonance in water is broadened if the molecular motions in water are reduced due to increasing viscosity or absorption of water on large molecules. Small displacements of the resonance lines (shifts) may also occur if the chemical bonding of protons is changed.

The proton magnetic resonance method, when applied on biological samples, primarily gives information about the nature of the water in the tissues, because water is their main compo-

nent. The water present in tissues has, however, a structure different from that of "free" water because of the presence of electrolytes and macromolecules of different kinds (proteins, mucopolysaccharides, collagen, etc.) capable of binding water molecules to their surfaces. The total water content in the human gingiva is thought to be about 65 %. No information is available on the nature or strength of the water binding in the gingiva. This paper is a preliminary study on the state of water in normal and diseased gingiva.

MATERIAL AND METHODS

Pieces of clinically healthy and diseased gingiva were excised in connection with oral surgery. In all there were 13 samples. The tissue pieces weighed about 25 mg and were immediately introduced in small soda glass tubes and sealed with wax. These tubes were later directly inserted in the magnetic resonance spectrometer. Immediately after filling the tubes, they were put in a refrigerator and later in a freezing box (—23° C) for storage until the investigation. Before the magnetic resonance spectroscopy was performed, the samples were allowed to take on room temperature (+20° C) and were centrifuged down in the test tubes and inserted in the spectrometer.

After the spectroscopic investigation the samples were examined microscopically after staining according to *van Gieson* and with haematoxylin-eosin.

The equipment used for magnetic resonance has been described earlier (*Odeblad,* 1961). The magnetic field was 4930 gauss and the proton frequency 16.7 Mc/sec. The resolution was 0.2 ppm (3 c/sec) as indicated by the resolution of alcohol spectrum (Fig. 1). For the measurement of magnetic resonance line widths, the audiomodulation side bands described by *Arnold & Packard* (1953) were used. The side bands were introduced at 170 c/sec on each side of the sample resonance. The full line width at half intensity was measured with reference to the side bands (Fig. 2). From the value of the full line width (B) the spin-spin interaction time (T_2) was calculated according to the equation

$$T_2 = 1 / \pi \cdot B$$

Table 1

Magnetic resonance line widths and spin-spin interaction times for various gingival samples.

Case	Age	Clinical diagnosis	Sample location	Mean line width c/sec	S.D. of mean line width	Number of observations	T_2 millisec
			PAD* group N				
1a	39	Gingival tissue	Toothless area $\underline{7}\rfloor$	61	1.8	12	5
b		Gingival sulcus	Mesially $\underline{8}\rfloor$	37	2.3	8	9
c		Gingival sulcus	Distally $\underline{8}\rfloor$	48	6.0	4	6
2a	52	Pericoronal sac	Nonerupted tooth $\underline{3}\rfloor$	45	1.1	7	7
		Mean (weighted)		49	1.2	—	6.8
			PAD* group D				
2b		Periodontitis chron.	Interdentally $\underline{21}\rfloor$	43	1.0	5	7
c		Periodontitis chron.	Interdentally $\underline{21}\rfloor$	28	1.4	8	12
3a	64	Periodontitis chron.	Interdentally $\overline{1\,1}$	28	2.7	5	11
b		Periodontitis chron.	Interdentally $\overline{1\,1}$	31	2.5	8	10
c		Periodontitis chron.	Interdentally $\overline{1\,1}$	38	2.0	6	8
		Mean (weighted)		33	0.9	—	9.6
			PAD* group M				
3d		Periodontitis chron.	Interdentally $\overline{1\,1}$	42	3.7	7	8
4	43	Gingivitis chron.	Buccally $\overline{8}\rfloor$	36	1.3	16	9
5	53	Periodontitis chron.	Mesially $\lfloor\overline{7}$	30	0.7	4	11
6	31	Periocoronitis chron.	$\overline{8}\rfloor$	30	0.9	14	11
		Mean (weighted)		34	0.9	—	9.8

* The pathologico-anatomic diagnoses were classified in three groups: —

N. Includes only samples of tissue rich in collagen and with only a few cells.

D. Includes only samples of granulation tissue containing a loose connective tissue rich in cells and poor in collagen.

M. Includes samples with pieces of both collagen-rich tissues and granulation cell-rich tissues. There is a slight inflammation in parts of the specimens.

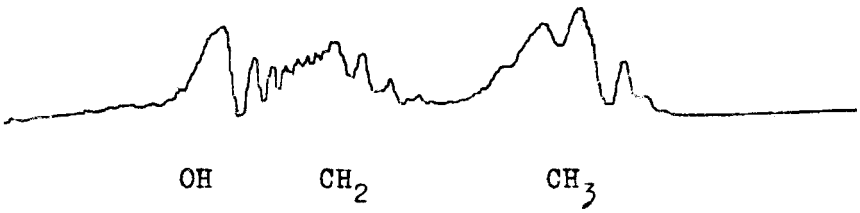

Fig. 1.

Proton resonance of ethyl alcohol showing the resolution of the spectrometer at spinning of the sample with a speed of about 60 rev./sec. The first order splitting is the chemical shift between OH, CH_2 and CH_3 groups.

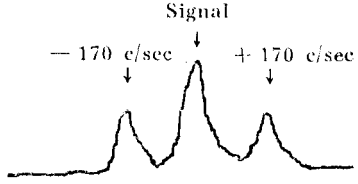

Fig. 2.

A typical proton resonance signal with the two sidebands at ± 170 c/sec (gingival tissue).

RESULTS

The results from the investigation are compiled in Table 1. From a pathologico-anatomic point of view the samples were classified in three groups, viz. (1) one (N) including only samples of tissue rich in collagen and with only a few cells, (2) one (D) including only samples of granulation tissue containing a loose connective tissue rich in cells and poor in collagen, and (3) one mixed group (M) where each sample includes pieces of both collagen-rich tissue and cell-rich granulation tissue. There is a slight inflammation in parts of the specimens.

The average values of resonance line widths were 48 c/sec for group N, 33 c/sec for group D and 34 c/sec for group M. The average values of spin-spin interaction time (T_2) were respectively 6.8, 9.6, and 9.8 millisec.

DISCUSSION

The values obtained for the line widths have to be discussed with reference to the state of water in the tissue. Pure water has a very small line width (about 0.2 c/sec) and a spin-spin interaction

time of about 2 sec. Our values indicate that the water phase in gingival tissue is markedly different from pure water.

The compounds which may be largely responsible for the binding of water in gingival tissue are collagen, mucopolysaccharides (ground substances) and cellular elements. The water binding capacity of each of these compounds depends on the surface exposed to water, on the number of hydrophile groups per unit of surface and, finally, on the strength of the bonds with water.

As many of these fundamental properties are unknown, a proper discussion of our results cannot be conducted. We expected, however, to find a much broader line in the collagen-rich samples than in the granulation tissue, because collagen may exhibit much less molecular motion than the macromolecules present in the ground substance. The explanation for our finding may be that collagen fibrils are arranged in such a way that a relatively small surface is exposed to the tissue water and consequently is less effective in tieing up water molecules.

Additional information on the so-named spin-lattice relaxation time of protons may also be of importance for understanding of the state of water. Work is therefore in progress to examine spin-lattice relaxation time (T_1) on different kinds of gingival samples as well as careful measurements on water contents in gingiva.

SUMMARY

The paper is a preliminary report on the nature and strength of the water binding in normal and diseased gingiva by means of nuclear magnetic resonance spectroscopy.

RÉSUMÉ

RÉSONANCE MAGNÉTIQUE DE PROTON DANS LE TISSU GINGIVAL HUMAIN

Cet article est le compte-rendu d'une étude préliminaire sur la nature de la puissance de la fixation de l'eau dans la gencive saine et dans la gencive malade, par spectroscopie de la résonance magnétique nucléaire.

ZUSAMMENFASSUNG

DIE PROTONMAGNETISCHE RESONANZ DES MENSCHLICHEN ZAHNFLEISCHES

EINE VORLÄUFIGE MITTEILUNG

Die vorliegende Arbeit ist eine vorläufige Mitteilung über das Wesen und die Stärke der Wasserbindung im normalen und pathologischen Zahnfleisch untersucht mittels kernmagnetischer Resonanzspektroskopie.

REFERENCES

Arnold, J. T. & M. E. Packard, 1951: Variation in absolute chemical shifts of hydroxyl groups of methyl and ethyl alcohols. J. chem. Phys. *19:* 1608.

Odeblad, E., 1961: A department for medical research with nuclear magnetic resonance. Acta Isotopica. *1:* 27.

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