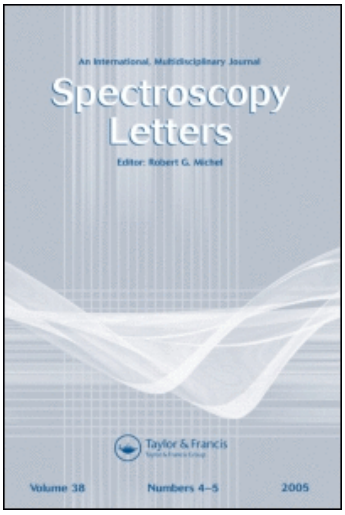

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A Comparative Study of Water T_1 and T_2 Nmr Relaxation Times in Healthy and Pathological Blood Fluid

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A COMPARATIVE STUDY OF WATER T_1 AND T_2 NMR RELAXATION TIMES IN HEALTHY AND PATHOLOGICAL BLOOD FLUID

KEY WORDS : NMR blood water T_1 and T_2 relaxation times , Macrocytic Anaemia , Chronic Lymphatic Leukaemia , Chronic Myeloid Leukaemia .

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ABSTRACT

Water protons T_1 and T_2 relaxation times in samples of whole blood , obtained from healthy people and from patients affected by Macrocytic Anemia on one side and Lymphatic and Myeloid Leukemia on the other , have been measured with the FT NMR technique at 80 Mhz and at 25 ^{0}C. No significant difference with respect to the value of the spin lattice relaxation time parameter measured for the healthy control group is experimentally evident in the case of the Macrocytic Anaemia while the spin spin relaxation time increases in magnitude. On the reverse both the leukemic cases present a significant ($p < 0.001$) increase in the relaxation times with respect to the control group. The experimental relaxation data belonging to the anaemic case show a linear correlation with the red cells volume while that obtained for the two leukaemic cases appear linearly

correlated with the total white cell numbers. From the relaxation data an estimate of the amount of water tightly bound to the white cells membrane can be determined which results roughly thirty times lower than that bound to the red cells membrane. In this work is also presented a step by step outline of the water relaxation behavior which starts with the pure water and ends with the water in the whole blood supported by relaxation experiments done on the isolated blood main components.

INTRODUCTION

Since the early applications of the NMR technique to biology, the determination of the water proton spins relaxation times in a variety of living tissues has marked a significant progress of the knowledge about their intrinsic properties.
Exhaustive reviews which deal with the many experiments performed by several authors on these topics have been compiled by Benè[1] and Bottomley[2] with a particular emphasis toward the applications of the relaxation parameters in the medical science MRI , directed to the early safe and quick detection of several diseases, e.g. cancer, involving living tissues, such as whole blood and other biological fluids.

The determination of both T_1 and T_2 in plasma and native blood in diverse experimental environments has stimulated a valuable theoretical improvement for the models suggested for explaining water relaxation time behavior in samples of healthy and diseased whole blood. The assumed models cover different approaches for the explanation of the blood water proton spin magnetic relaxivity. These models go from the bimodal distribution approach[3] of the blood water molecules rotational[4] and translational[5] correlation times, to the finite difference scheme adopted for the numerical solution of the average magnetization vector diffusion equation[6] . The vector diffusion equation explains the blood water bulk phase and the compartmented water inside the blood red cells with the resultant spatial coordinates dependence of the transversal T_2 relaxation time. Despite the different models used , the experimentally determined blood water relaxation times are similar in each published work. I will discuss more on these arguments in the rest of this paper. One of the first authors to report an increased variation between the water relaxation times in some forms of Leukemia and healthy human plasma has been McLachlan[7] , but his conclusions were only indicative of the effect.

In this work I will report the results of T_1 and T_2 measurements on a significant pool of whole blood samples in 50 subjects of a healthy control group, 34 affected by Macrocytic Anemia (**MA**), and 40 and 30 affected by Lymphatic and Myeloid Leukemia (**CLL, CML**), respectively. The results show that the average blood water relaxivity data for the **MA** case does not differ significatively from that obtained for the healthy control group , while a non negligible difference is evident for the **CLL** and **CML** cases. Furthermore, the statistical treatment of the experimental relaxation data with the blood parameters obtained from the clinical analysis sheets gives evidence for a linear correlation with the average red cells volume for the **MA** case and with the total white cells number for both the **CLL** and **CML** cases.

Also in this paper a short outline is presented of the T_1 and T_2 water proton dependence from the solution composition which starts from the pure state up to the whole blood water using the recent relaxation theories and supported by measurements done on properly prepared water suspensions of the isolated main blood components.

EXPERIMENTAL SECTION

Albumine , α globuline, and fibrinogen all products derived from human blood were purchased from Fluka Chemicals. Liophylized ram erythrocites membrane ghosts powder, labeled ProRubenosticon , was a product purchased from Organon TeKnica. Solutions of the different blood protein components were prepared at a concentration nearly equal to that of human plasma , i.e. 10 and 90 % weight in total proteins and water at STP equilibrium. Tris buffer was added to reach the physiologic pH. Heparinized whole blood samples for all the people involved in this work were anonimously obtained from the Clinical Blood Analysis Service of the Padua University. All the samples went with the relative blood analysis parameters sheets which were successively used for the statistical correlations. Several diverse weight to weight artificial plasma / ram erythrocytes membrane ghosts samples were prepared , maintaining constant erythrocyte weights and linearly changing the weight of the added plasma to reach the normal average ratio of the native blood, i.e., 44 and 56 % weight in wet red cells membrane and plasma.

At low plasma / erythrocytes ghost ratios, the samples were all prepared in sealed 3 mm o.d. capillaries, 15 mm long. These were stored for several hours in a box at a temperature of 15 ^{0}C in order to reach the complete water diffusion equilibrium. These were checked with a fast T_1 determination , i. e. null point method , up to the time of no detection of change in the longitudinal water relaxation parameter. The T_1 and T_2 determinations of the whole blood and red cell membrane ghosts suspensions in plasma were completed in 3 mm o.d. sealed capillaries fitted with Teflon plugs into standard 5 mm NMR tubes filled externally with deuterated water, 99.99 % in Deuterium content , for the spectrometer lock. The spectrometer used in this work was a Bruker WP80SY FT instrument at the proton frequency of 80 Mhz and with the probe controlled at the temperature of 25 ± 1 ^{0}C.

Standard relaxation time detection techniques were used[8], i.e., the Inversion Recovery for the T_1 experiments and the Hahn method as modified by Carr , Purcell, Meiboom , Gill for the T_2 determinations. The experimental parameters for the observation of the water resonance were 240 Hz for the Sweep Width, 0.03 Hz/Pn for the Digital Resolution , π RF Pulse Width equal to 9.4 μs and Time Delay list optimized to cover all the time domain of the single exponential decay . Each measurement was repeated at least three times in succession and the results averaged with a root mean square error of ± 5 %, which rises to 10 % in the case of low S/N ratio samples . The relaxation times data are reported in the Tables 1 - 4 with the comments added when pertinent.

RESULTS AND DISCUSSION

A) Proton relaxation times behavior from the pure water up to whole blood in healthy people.

Table 1 summarizes the water protons relaxation times T_1 and T_2 measured in this work and partly described in the literature[9,10] , arranged in decreasing order from ultrapure water up to the mix plasma / ram erythrocyte membrane ghosts powder. As may be noted, the ultrapure water has the highest proton relaxation times , whose aspects have been discussed extensively by Krycnicki[9]. The noticeable drop in the relaxation times of the

Table 1. Water T_1 and T_2 relaxation times of diverse biological solutes at 25 °C .

	$\frac{x}{1-x}$ [a]	$<T_1>$ (s)	$<T_2>$ (s)	$\Delta\nu_{1/2}$ (Hz)	T_2^* [b] $= [\pi(\Delta\nu_{1/2} - \Delta\nu^0_{1/2})]^{-1}$ (s)
Ultrapure		3.57±0.07	3.60±0.07		
Undegassed [c]		2.56±0.03	1.47±0.08	0.7	
Human plasma [d]		1.98±0.04	0.46±0.05	1.5	0.40
Artificial plasma [e] **with 0.04 Albumine , 0.024 Globuline , 0.035 Fibrinogen in %**		1.81±0.05	0.32±0.06	2.0	0.25
Whole blood [d]		1.14±0.11	0.07±0.02	8.0	0.043
Suspension of liophylized ram erythrocytes in plasma					
	4.882	0.11±0.02	0.014±0.008	28.5	0.012
	2.448	0.22±0.03	0.030±0.007	8.4	0.038
	1.632	0.31±0.05	0.040±0.014	7.3	0.044
	1.250	0.39±0.07	0.054±0.025	5.8	0.055
	1.000	0.47±0.05	0.063±0.010	4.6	0.070
	0.334	0.93±0.07	0.153±0.025	2.2	0.145
*	0.250	1.10±0.04	0.180±0.030	1.7	0.188
*	0.200	1.20±0.05	0.200±0.035	1.5	0.212

Notes : (a) $\frac{x}{1-x}$ is defined as [gram weight fraction of ram erythrocytes / gram weight fraction of plasma] [4].

(b) Equation valid for $T_2 < 1$ s : see Reference 8. $\Delta\nu^0_{1/2} = 0.7$ Hz refers to the pure water linewidth assumed as instrumental magnetic field inhomogeneity.

(c) Average over 20 different measures.

(d) Average over 50 samples.

(e) The solute concentrations are given in [gram weight of proteins /100 ml of water]. Average over 10 measures.

Table 2. Water T_1 and T_2 relaxation times of whole blood in subjects affected by Macrocytic Anaemia (MA) at 25 °C .

#	Number of cases	$<T_1>^{(a)}$ (s)	$<T_2>^{(a)}$ (s)	$\Delta\nu_{1/2}$ (Hz)	$T_2^{*\,(b)} = [\pi(\Delta\nu_{1/2} - \Delta\nu^0_{1/2})]^{-1}$ (s)	$MCV^{(a)}$ (fL)
1	4	0.90±0.03	0.09±0.03	4.4	0.08	108.1
2	3	0.95±0.03	0.10±0.04	4.3	0.09	107.6
3	5	0.98±0.04	0.11±0.02	3.4	0.12	106.9
4	2	0.99±0.04	0.10±0.05	3.0	0.14	104.2
5	2	1.15±0.08	0.12±0.04	3.0	0.15	102.3
6	5	1.18±0.05	0.13±0.03	2.9	0.15	101.8
7	3	1.30±0.05	0.12±0.02	2.3	0.21	100.8
8	5	1.37±0.04	0.12±0.04	2.0	0.27	99.5
9	5	1.62±0.07	0.14±0.05	2.0	0.27	91.5
	Total= 34	$T_{1\,av}$ = 1.16 ±0.22	$T_{2\,av}$ = 0.11 ±0.02	$\Delta\nu_{1/2av}$ = 3.0 ±0.84	$T_{2\,av}^{*}$ = 0.11 ±0.07	MCV_{av} = 103 ±4.9

Notes : (a) Linear regression : $<T_1> = 5.71 - 4.5\ 10^{-1}$ MCV with | R | = 0.972 and
$<T_2> = 0.37 - 2.5\ 10^{-3}$ MCV with | R | = 0.890.
MCV = Medium Corpuscle Volume in femtoLitres.

undegassed water in STP equilibrium has been discussed[10] in term of the paramagnetic effect of the dissolved oxygen with two umpaired electron spins in the ground state. The time averaged electron spin magnetic moment of 1.2 Bohr magnetons which can be extracted from the relaxation times data reported in this work agrees tightly with the value reported in previous literature[10].

It is interesting to note the aspect of the water proton relaxation times in human plasma with the average composition respectively of 90 and 10 % weight of water and different proteins. These proteins include albumin fractions , diverse species of globulins, fibrinogen and other small molecules. The previous study of McLachlan[7] at 20 Mhz reports T_1 and T_2 values of 1.48 s and 0.57 s, respectively, for healthy people. The data for the human plasma obtained in this work at 80 Mhz, averaged over 50 different samples , are consistent with that previously published[7] taking in account the linear relationship between the relaxation rates and the actual experimental proton frequency[5].

Moreover the water proton relaxation times obtained for the artificial plasma prepared by dissolving the principal proteins in water at physiologic pH, and reported in

Table 3. Water T_1 and T_2 relaxation times of whole blood in subjects affected by Chronic Lymphatic Leukaemia (CLL) at 25 °C .

#	Number of cases	$<T_1>^{(a)}$ (s)	$<T_2>^{(a)}$ (s)	$\Delta\nu_{1/2}$ (Hz)	$T_2^{*(b)} = [\pi(\Delta\nu_{1/2} - \Delta\nu^0_{1/2})]^{-1}$ (s)	$WBC^{(a)}$ ($10^3/mm^3$)
1	5	1.26±0.02	0.09±0.05	5.3	0.07	60.5
2	8	1.29±0.04	0.09±0.02	3.9	0.10	60.0
3	3	1.30±0.04	0.13±0.03	3.5	0.11	53.3
4	4	1.37±0.05	0.12±0.04	3.2	0.13	48.0
5	4	1.44±0.08	0.13±0.03	3.0	0.15	46.0
6	6	1.52±0.05	0.14±0.04	2.8	0.15	45.6
7	3	1.55±0.07	0.15±0.02	2.8	0.15	24.9
8	4	1.57±0.04	0.15±0.05	2.5	0.18	19.4
9	3	1.65±0.07	0.16±0.04	2.3	0.21	14.4
	Total= 40	$T_{1\,av}$ = 1.44 ±0.13	$T_{2\,av}$ = 0.14 ±0.03	$\Delta\nu_{1/2av}$ = 3.4 ±0.80	$T_{2\,av}^{*}$ = 0.14 ±0.07	WBC_{av} = 42 ±16

Notes : (a) Linear regression : $<T_1> = 1.74 - 7.2\ 10^{-3}$ **WBC** with | R | = 0.940 and
$<T_2> = 0.18 - 1.3\ 10^{-3}$ **WBC** with | R | = 0.890.
WBC = Total White Blood Cells number.

Table 1, give values which are strictly comparable to that for the native plasma. A further decrease in water proton relaxation times, particularly evident for T_2 , is observed for the whole blood samples in the healthy control group. In fact, the averaged T_1 and T_2 values are respectively 1.14 s and 0.07 s. The same table reports the T^*_2 values determined from the width of the water magnetic resonance lineshape for sake of comparison with the natural transverse relaxation time measured with pulsed methods. It can be observed that when the condition[8] $T_2^{-1} >> \gamma\,\Delta B/\,2\pi$, where ΔB represent the magnetic field inhomogeneity, is satisfied the two methods of measurement provide nearly equal values. The problem of the factors involved about the microscopic origin of the relaxation times for the water in the whole blood samples has been faced with different aprroaches by several authors with outstanding contributions given by Zipp[3] , Finch[4] and more recently by Santyr[6] and Ceckler[11] et al.

Ceckler[11] et al. Worked with human erythrocytes membrane ghosts derived from disrupted native blood red cells, and with the use of the two sites fast exchange formalism

Table 4. Water T_1 and T_2 relaxation times of whole blood in subjects affected by Chronic Myeloid Leukaemia (CML) at 25 °C .

#	Number of cases	$<T_1>^{(a)}$ (s)	$<T_2>^{(a)}$ (s)	$\Delta\nu_{1/2}$ (Hz)	$T_2^{*(b)} = [\pi(\Delta\nu_{1/2} - \Delta\nu^0_{1/2})]^{-1}$ (s)	$WBC^{(a)}$ (10^3 /mm^3)
1	4	1.22±0.04	0.08±0.03	4.5	0.09	150
2	6	1.35±0.05	0.08±0.02	4.0	0.10	66
3	7	1.38±0.06	0.09±0.03	3.9	0.10	61
4	5	1.50±0.05	0.09±0.02	3.5	0.12	38
5	8	1.55±0.08	0.10±0.03	3.4	0.12	21
	Total= 30	$T_{1\,av}$ = 1.40 ±0.12	$T_{2\,av}$ = 0.09 ±0.03	$\Delta\nu_{1/2av}$ = 3.9 ±0.40	$T_{2\,av}^*$ = 0.11 ±0.03	WBC_{av} = 67 ±44

Notes : (a) Linear regression : $<T_1> = 1.56 - 2.3\ 10^{-3}$ **WBC** with | R | = 0.950 and
$<T_2> = 0.09 - 9.8\ 10^{-3}$ **WBC** with | R | = 0.780.
WBC = Total White Blood Cells number.

between the free water in the plasma phase and the tightly bound to red cells membrane water phase. This work has found that the T_1 for membrane bound water amounts to 0.092 s at the value of the c parameter , defined as the ratio : g bound water / g of solid membrane ghosts , and is equal to 0.28 at the proton resonance frequency of 24 Mhz and 30 °C.

In this work it have followed a similar approach by measuring the water protons relaxation times for a series of suspensions of liophylized ram erythrocyte membrane ghosts powder at constant weight mixed with variable amounts of artificial plasma in which the parameter $\frac{x}{1-x}$, defined in Equation [1], changes between 0.2 up to 5.
The resulting T_1 and T_2 of these mixes are also reported in Table 1 together with the T^*_2 derived from the water resonance lineshape. The analysis of the experimental data has been done by using the fast two sites exchange relationship[4]

$$\frac{1}{T_i} = \left\{ c\left[\frac{1}{T_b} - \frac{1}{T_f}\right]\right\} \frac{x}{1-x} + \frac{1}{T_{if}} \qquad [1]$$

where i = 1, 2 , the indexes b , f refer to the bound and free water , c has been previously defined and x , 1 - x are in order the weight fractions of the solid ram erythrocyte membrane ghosts and artificial plasma water.

The linear relationship **[1]** accounts quite well for the experimental T_1 of the red cell membrane ghosts suspensions in plasma by fitting to a straight line with slope 1.74. From the slope, using for T_1 the limit experimental value of 0.113 s , which compares well with the values reported in the literature[3, 4, 5] obtained with a variety of experimental techniques , I obtain a value for **c** of 0.21 at the proton resonance frequency of 80 Mhz and at 25 °C. It is worth noting that the assumption[4, 5] to neglect in the relationship **[1]** the erythrocyte membrane bound water residence time τ_r which has been recently determined[12] to have the value 0.013 s , nine times shorter than T_{1b} and hence negligible. Table 1 shows the T_1 data marked with * at the $\frac{x}{1-x}$ values in the interval between 0.25 ans 0.2 are similar to the average whole blood T_1 in the healthy control group and this fact gives solid evidence that the native blood system can be treated safely with the two sites exchange theory[13] which is the simplest step to tackle more complex exchange features in living structures[6].

A simple calculation , done on the basis of the blood analysis clinical sheets , proves that for the healthy whole blood with density 1.1 g per ml , free plasma water 0.603 g and hematocrit 0.414 g , the inside red cell water amounts to 0.278 g from which the ratio $\frac{x}{1-x}$ equal to 0.23 is easily determined.

The transversal relaxation time in the same erythrocytes membrane ghosts / plasma system can be determined from the slope of the linear plot of T_2^{-1} vs $\frac{x}{1-x}$. The value of the **c** parameter is 0.22, and the limit T_{2b} value is 0.015 s for water totally bound to the red cell membrane. The same value T^*_2 is obtained from the inspection of the proton water resonance linewidths. This experimentally determined transverse relaxation time agrees with the value of 0.010 s extrapolated at 25 °C from the low temperature T_2 reported by Zipp et al.[3] , but disagrees from the value of 0.130 s found by Santyr et al.[6] at 25 Mhz for *the spin-spin relaxation time for the water inside the blood red cells.*

The proton water T_2 measured inside the intact red cell compartments is very different from the T_{2b} which refers to the tightly membrane bound water protons and seems more similar to the value of 0.013 s reported by Lahajnar[12] for the water diffusional time through red cells membrane. Evidently in the context of the disrupted membrane state , hence lack of water diffusion across the intact red cell walls, this last diffusion time assumes the role of the water tightly bound residence time as described by Finch et al.[4] Such low transversal relaxation times are not unusual for water studied in lipidic molecular systems which mimic the membrane properties as demonstrated by Cecklar et al.[11] His work found in lipidic bilayer dispersions a range of water T_2 which spans the interval from 0.011 s in the phosphatidylglycerol / water system to 0.048 s in the egg phosphatidylcholine : cholesterol / water system.

B) Proton relaxation times behavior of water in selected blood pathologies.

Tables 2 , 3 , 4 , display the results of the measurements of the water T_1 , T_2 , T^*_2 for some important blood pathologies , i.e. Macrocytic Anaemia (**MA**), Chronic Lymphatic (**CLL**), and Myeloid Laeukemia (**CML**). The data are arranged as average over the number of cases which produce the same values within the constraint of the experimental error. The first blood pathology , **MA** , is characterized by a severe reduction of the blood red cells average number compared to healthy people. This blood property is combined with an abnormal increase in the red cells average size **MCV** and both factors are

responsible of the macroscopic feature, typical of this disease , called hyperchromism. Both these factors consistently reduce the blood's essential task to provide oxygen fuel to the cellular metabolism and to drain off the cellular waste products. THis condition results in severe damage to the living tissues and organism.

The two other blood diseases , **CLL** and **CML** , are distinguished primary by an unusual increase in the total blood white cell number **WBC** which sometime reaches a maximum peak of 2000 % higher than the average value of 7800 per mm^3 found in healthy blood, and as reported in the tables 3 and 4. The white cell total number hyperthrophism originates in the lymphatic network where a variety of white cells , the lymphocites , are produced; or in the bone marrow system where the rest of the blood white cells have their birthplace.

One of the first authors to report measurable differences in leukemic blood water proton relaxation times has been McLachlan[7] , who observes at 20 Mhz and 25 °C an increase of 6 % in both T_1 and T_2 with respect to healthy blood samples. The blood water relaxation time data obtained in this work show that the difference is negligible for T_1 , while a 63 % increase is displayed for T_2 for the case of **MA** affected patients when compared to relaxation times for healthy blood specimens. The corresponding differences in the blood water relaxation times for the leukemic cases confirms the McLachlan[7] observations. For the **CLL** and **CML** cases, the percent differences are 27 and 23 for the spin lattice and 100 and 25 for the transversal relaxation times as compared to the values obtained for the healthy subjects control group.

This work shows a correlation for the diseased blood water proton relaxation times to the corresponding clinical blood analysis multiparameters. The linear regression correlation is as $| R | > 0.78$, with the average red cells volume number **MCV** for the **MA** case, and with the total average white cells number **WCB** for the **CLL** and **CML** cases. The linear regression coefficients are reported in the footnotes of the pertinent tables.

It is not an easy task to determine the origin of the variation in water proton relaxation times between the diseased and healthy blood, using the simple two site exchange kinetics model assumed previously in the discussion of the relaxation behavior of the blood in healthy people. The intrinsic deviation factors can go from either an altered distribution of the water rotational and translational correlation times [3,4] to an increased permeability of the diseased blood red cell membrane[5,6] due to any structural surfacial change[11] , which tend to equalize the relaxation times of the red cell internal water to that of the outside plasmatic water[6]. Using an empiric viewpoint, one can define the average volume ratio parameter as

$$v = \frac{\textbf{Red cells total volume}}{\textbf{White cells + Platelets total volume}} \qquad [2]$$

where the blood figurate corpuscles total volume is given in μ^3 for ml of whole blood. It is then possible to derive the relaxation equation for the case of three fast exchanging sites, labeled a for the water bound to the red cells, b for the water bound to the white and the platelets cells and f for the plasma water, in function of the variable v

$$\frac{1}{T_i} = v\frac{c_b x_b}{1-x_a-x_b}\left[\frac{1}{T_{ia}} - \frac{1}{T_{if}}\right] + \frac{c_b x_b}{1-x_a-x_b}\left[\frac{1}{T_{ib}} - \frac{1}{T_{if}}\right] + \frac{1}{T_{if}} \qquad [3]$$

which , for T_{ia} assumed nearly equal to T_{ib} , takes the form of a linear function vs

$[v + 1]$, with slope equal to $\frac{c_b x_b}{1 - x_a - x_b}\left[\frac{1}{T_{1a}} - \frac{1}{T_{1f}}\right]$, and intercept equal to $\frac{1}{T_{1f}}$.

The values of the v parameter for the healthy and pathologic blood , derived from the clinical blood analysis data sheets , are in order 51 and 47 for healthy and **MA** affected blood , and 4.5 and 11 for **CLL** and **CML** affected blood samples, respectively. The plot of the average relaxation rates for the diverse blood conditions vs $[v + 1]$ is linear for the spin lattice relaxation rate using the empirical equation as $\langle T_1^{-1} \rangle = 0.67 + 4 \cdot 10^{-3} [v + 1]$ ($R = 0.998$) and strongly non linear ($R = 0.587$) for the spin spin relaxation rate $\langle T_2^{-1} \rangle$.

From the T_{1a} and T_{1f} data in Table 1 and from the blood analysis data sheets one can calculate the value of 0.13 for the $\frac{x_b}{1 - x_a - x_b}$ parameter. Thus obtaining for c_b a value of 0.006. This number represents the ratio between the amount of the water (in g) tightly bound to each g of total white and platelet cells. This value is thirty times lower than the amount of water tightly bound to the red cell membrane. The linear behavior of the longitudinal relaxation rate and the non-linear result in the transversal relaxation rate suggest that the origin of the differences measured between the relaxivities, in the normal and pathologic blood points, are due to a decreasing weight of the secular term [3,4,13] in the theoretic relationship for T_2^{-1} . Thus the static component along the z axis of the random modulated $\langle B_l^2 \rangle$ local magnetic fields, with the parameter v, is related to the local magnetic field inhomogeneities of the actual blood sample. This factor acts in such a way that at different spin lattice relaxation times, the spin-spin relaxation times of the water tightly bound to the diverse cell membranes in the a and b phases cannot be assumed equal for different blood samples (with differing cellular composition), but changes intrinsically with the parameter v.

This topic has been discussed in detail elsewhere[6] when the simple two site exchange approximation breaks down either for a sizeable reduction in cell permeability (presumably this does not apply to the systems studied in this work), and / or when the cellular compartments which form the whole structure of the living tissue present strongly balanced different sizes.

I propose that this last qualitative explanation can apply to the cases of the **CLL** and **CML** affected blood in which the total volume of the white cell and platelet cell subsystems is nearly balanced with that of the red cells. This condition is indicated by a low value of the parameter v. In the the healthy and **MA** affected blood this same parameter increases greatly in magnitude with the result of a strongly unbalanced system. The study indicates that the water magnetic relaxivity in healthy and diseased whole blood is that of a two water phase system , as in the discrete[4,5] or continuous approach[6] . This relationship is valid for the condition where the red cell system overwhelms any other kind of cellular system (as indicated by high values of the parameter v). Conversely, in systems with a low value of the parameter v, as occuring in leukaemic blood samples, a more general water exchange theory applies satisfying the requirement for decreasing random relaxing magnetic fields.

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