OZONE THERAPY IN PATIENTS WITH VIRAL HEPATITIS "C" A CLINICAL STUDY

By

Prof. Dr. Mohamed Nabil Mawsouf*, **, Dr. Taha Tarek Tanbouli**& Dr.Wagdy Ibrahim El-Tayar**

** Cancer Institute, Cairo University*
*** Cairo Medical Center*

Abstract:

Hepatitis "C" is a medical problem in Egypt. The usual line of treatment is very expensive with major side effects and low efficacy especially in type 4, which is common in Egypt. The aim of this study is to evaluate the role of ozone as a safe line of treatment. This study included 60 type 4 hepatitis "C" patients, 45 males and 15 females. Their age ranged between 34 and 65 years. Investigations including C.B.C., liver function tests, A.F.P., serological tests for Bilharziasis, P.C.R. quantitative for H.C.V., prothrombin time and concentration and abdominal ultrasonography were done before and 8 weeks, 24 weeks after treatment with ozone. Patients received combined treatment of Major AutoHaemotherapy in a dose range from 4mg to 9 mg and rectal insuflation in a dose range from 6mg to 14 mg per visit. The numbers of visits were three times per week for 8 weeks followed by twice per week for 16 weeks. The general condition in 95% of cases improved. There was a decrease in the quantitative P.C.R. (viral load) in 91.67% of cases that reached –ve P.C.R. in 20 % of cases after 8 weeks treatment. The number of –ve P.C.R. cases for HCV virus increased to reach 36.67 % of cases after 24 weeks treatment. Ozone therapy was found to be an effective, safe and less expensive method in Hepatitis "C" patients.

Aim of the Study

This study was made to evaluate the effectiveness of ozone therapy in hepatitis C genotype 4 infections and to evaluate a proposed ozone therapy protocol in HCV genotype 4 treatment. This is a provisional study to be followed by another study.

Introduction

Hepatitis C (HCV) is a worldwide medical problem. It is estimated that more than 300 millions on earth are suffering from HCV. Hepatitis C is a major medical problem in Egypt. It is postulated that more than 15% i.e. more than 10 millions of the population in Egypt are suffering from HCV. This disease is slowly progressing, detected mainly accidentally, devitalizing and difficult to treat. The usual line of treatment is very expensive with major side effects and low efficacy.

HCV in most cases leads to complications e.g. liver cirrhosis, ascitis, liver carcinoma and ultimately liver cell failure. Liver Cirrhosis is estimated to develop in 20 -25 % of patients with HCV within 20 years. Hepato-cellular carcinoma in 5% of patients.

It is not only a medical problem, but also an economic problem (less work, less production and very high costs of usual treatment).

So far there are six genotypes of HCV with worldwide prevalence of genotypes 1,2 &3. In Africa genotype 4 and 5 are more dominant. In Asia genotype 6 is more dominant. Genotype differences have shown varying susceptibility to antiviral therapy. In Egypt genotype 4 is prevalent and it is known that is relatively resistant to antiviral treatment.

The main line of treatment nowadays for hepatitis C includes interferon and ribavirin. Ribavirin and interferon have significant medical and psychiatric side effects.

Antiviral effect of ozone

Ozone is a powerful oxidizing agent. It disrupts viral envelope proteins, lipoproteins, lipids, and glycoproteins. The presence of numerous double bonds in these unsaturated molecules makes them vulnerable to the oxidizing effects of ozone. Molecular architecture is disrupted and widespread breakage of the envelope ensues. Deprived of an envelope, virions cannot sustain nor replicate themselves. Ozone proper, and the peroxide compounds it creates, may directly alter structures on the viral envelope, which are necessary for attachment to host cells. Peplomers, the viral glycoproteins protuberances that connect to host cell receptors are likely sites of ozone action. Alteration in peplomer integrity impairs attachment to host cellular membranes foiling viral attachment and penetration.

Ozone stimulate leucocytes function and cytokine production

Ozone is a powerful oxidant by itself and leads to production of peroxides with an oxidative power. H2O2 crosses the cell membrane and activates the cytoplasmic gene-regulatory nuclear factor kappa B, ultimately causing the transcription of mRNAs of several cytokines, namely interleukin (IL-1,2,4,6,8,10), tumor necrosis factor (TNF-$\propto$) and interferon (IFN β and γ).

In HCV, viral load appears to be a major factor in the invasiveness and virulence of the disease process. Ozone induces the release of cytokines by leucocytes. Stimulation of the immune mechanisms will lead to significant reduction of circulating virions.

Patients and methods

This study included 60 type 4 hepatitis "C" patients, 45 males and 15 females. Their age ranged between 34 and 65 years. Investigations including C.B.C., liver function tests, A.F.P., serological tests for Bilharziasis, P.C.R. quantitative for H.C.V., prothrombin time and concentration and abdominal ultrasonography were done before and 8 weeks, 24 weeks after treatment with ozone. Patients received combined treatment of Major AutoHaemotherapy in a dose range from 4mg to 9 mg and rectal insuflation in a dose range from 6mg to 14 mg per visit. The numbers of visits were three times per week for eight weeks followed by twice per week for sixteen weeks.

Investigations were repeated after 8 and 24 weeks of treatment (but in this study we are focusing on PCR quantitative and Liver enzymes)
General health and daily activity were observed.

Ozone Treatment Protocol

First session of major AutoHaemotherapy was given in a concentration of 25µ/ml ozone in oxygen for two successive times, then increased to 30µ/ml ozone in oxygen for another two successive times and so on, increasing the concentration by 5µ/ml every two sessions till reaching a maximum of 60µ/ml were this concentration was fixed till the end of treatment course. The rationale of start low and go slow was respected. The ozone in oxygen volume was fixed in all sessions at 150 ml. The blood weight was constant in each session at 150 gm.
First session of rectal insufflation was given in a concentration of 20µ/ml ozone in oxygen with a volume of 300 ml for two successive times, then increased to 25µ/ml ozone in oxygen with the same volume for another two successive times, then 30µ/ml x 300 ml twice, then 35µ/ml x 300 ml twice followed by 35µ/ml x 350 ml twice till we reach a maximum of 40µ/ml x 350 ml were this concentration and volume was fixed till the end of treatment..

Results

It was found that following eight weeks of ozone therapy, the viral load decreased in 91.67% of cases (P value < 0.001) that reached zero level in 20 % 0f cases. Following 24 weeks of ozone therapy, there was further decrease of the viral load that reached 95 % of cases (P value < 0.001) with a zero level in 36.67 % 0f cases. After eight weeks of ozone therapy, the abnormal enzyme levels were back to normal in 20 % of cases (P value < 0.001) for the SGPT enzyme, and were back to normal in 23.33 % of cases (P value < 0.013) for the SGOT enzyme (normal levels are < 42 U/L for the SGPT enzyme, and < 45 U/L for the SGOT enzyme).

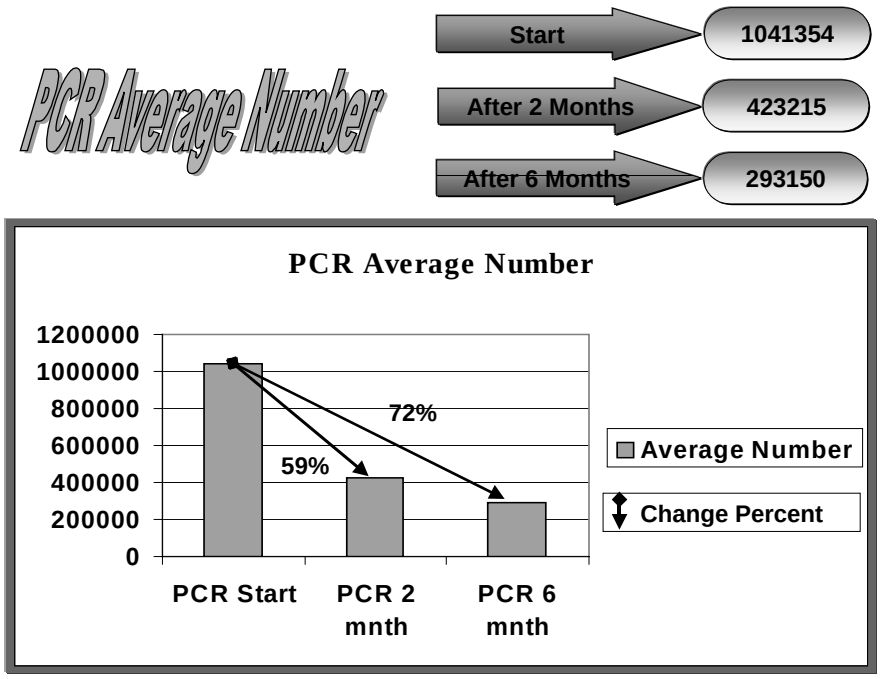
Table 1

Serial Number	Sex	PCR	PCR 2mnth	PCR 6 mnth	SGOT	SGOT 2 mnth	SGPT	SGPT 2 mnth
1	Male	89000	38000	25000	32	37	36	36
2	Male	65000	41000	0	29	23	21	20
3	Male	86000	31000	9300	56	40	50	39
4	Male	2800	1100	9000	67	40	51	37
5	Male	6054000	0	0	78	46	43	38
6	Male	892169	516000	30000	32	31	36	32
7	Male	200000	74000	18000	36	35	38	38
8	Male	49600	0	0	33	32	33	33
9	Male	71500	20000	0	37	37	36	35
10	Male	200000	60000	23000	38	33	32	32
11	Male	57000	22000	10000	76	45	58	40
12	Male	12000	0	0	41	40	27	37
13	Male	78000	31000	0	42	39	36	36
14	Female	87360	18800	0	34	34	12	26
15	Female	6800	0	0	41	31	52	33
16	Female	40000	15000	8000	15	8	14	6

Serial Number	Sex	PCR	PCR 2mnth	PCR 6 mnth	SGOT	SGOT 2 mnth	SGPT	SGPT 2 mnth
17	Male	7000	4200	3000	57	17	92	25
18	Male	2726000	2814000	2519000	67	39	43	33
19	Male	11650	9850	8500	39	37	33	32
20	Male	410800	290600	150000	37	28	13	15
21	Male	96000	0	0	90	60	95	67
22	Female	200000	6000	2500	70	57	50	40
23	Male	96000	23700	11500	26	37	22	21
24	Male	56000	30000	29000	63	40	31	31
25	Female	93000	40300	16500	33	51	34	45
26	Female	56000	8600	0	37	30	32	28
27	Male	1779992	123000	54000	40	40	37	36
28	Male	29000	0	0	36	22	33	18
29	Female	3372000	127000	42000	47	41	42	36
30	Female	648931	792000	687000	56	42	48	41
31	Male	2000	0	0	38	37	33	32
32	Female	9000	0	0	39	38	37	32
33	Male	7500000	500	0	69	74	130	57
34	Female	31500	21000	9500	57	40	47	32
35	Male	113000	45000	32000	92	62	72	46
36	Male	45160	21211	7500	39	38	34	34
37	Male	16208400	15392000	12574000	37	36	33	33
38	Male	8000	6500	5200	36	36	33	32
39	Male	90000	38000	12300	39	46	54	38
40	Male	186000	0	0	39	37	37	32
41	Male	75000	19200	4000	67	37	39	32
42	Male	70000	18000	5300	36	35	33	32
43	Male	960000	670000	330000	37	36	14	11
44	Male	41592	0	0	72	76	88	111
45	Male	1800000	20000	3200	66	44	50	39
46	Male	30000	30000	28000	315	45	124	39
47	Male	27000	3000	1200	47	42	39	32
48	Male	78000	40000	30372	249	195	372	176
49	Male	220000	823647	0	12	12	11	10
50	Female	48000	1100	0	20	20	26	26
51	Male	2912000	1322000	0	39	37	33	32
52	Female	187000	244000	198000	131	69	80	50
53	Male	20000	7600	1600	8	83	37	31
54	Male	823000	0	0	142	60	168	70
55	Female	1080000	760000	520000	66	35	55	42
56	Male	5357000	6000	0	64	39	122	60
57	Female	6000	0	0	43	39	40	32
58	Male	5200000	624000	112000	38	34	42	22
59	Male	1700000	83000	19500	21	12	23	11
60	Female	80000	60000	40000	44	40	35	35

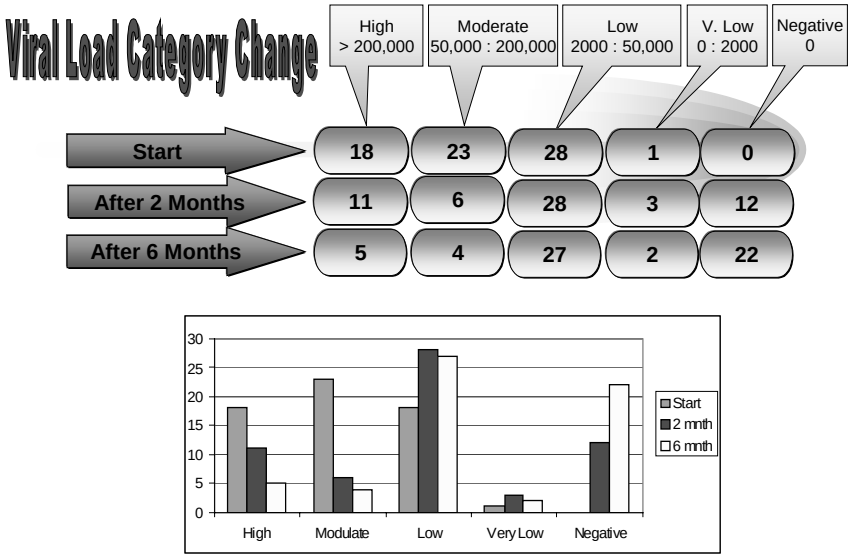
PCR and enzyme levels in all patients before and after ozone therapy

Fig. 1



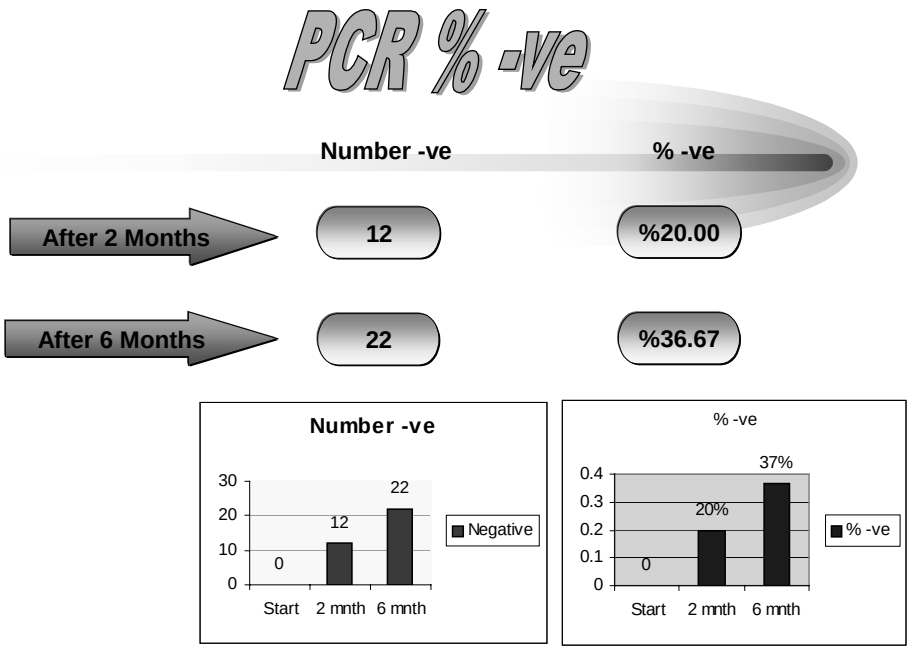
PCR average number before, during and after ozone therapy

Fig. 2



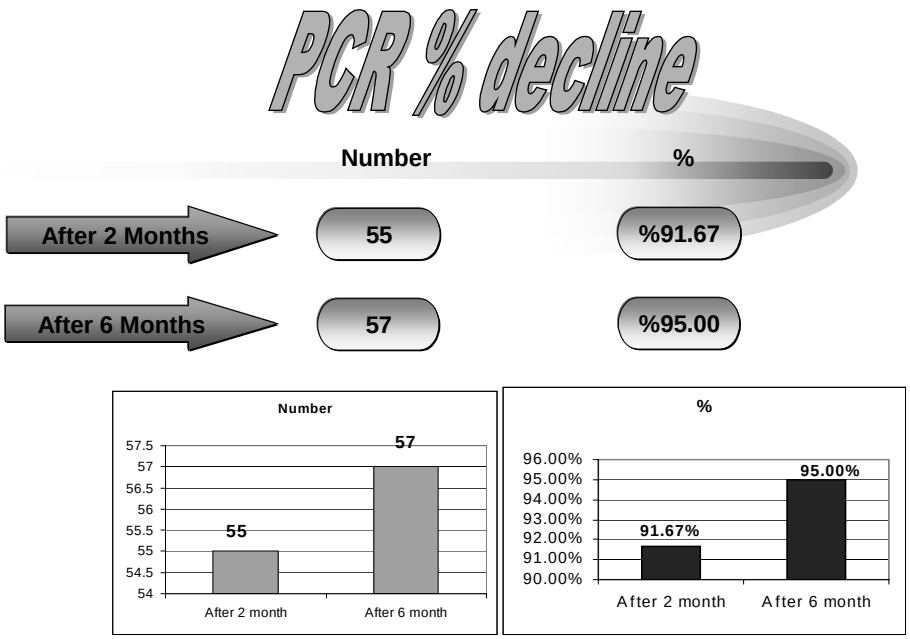
Viral load category numbers before, during and after ozone therapy

Fig. 3



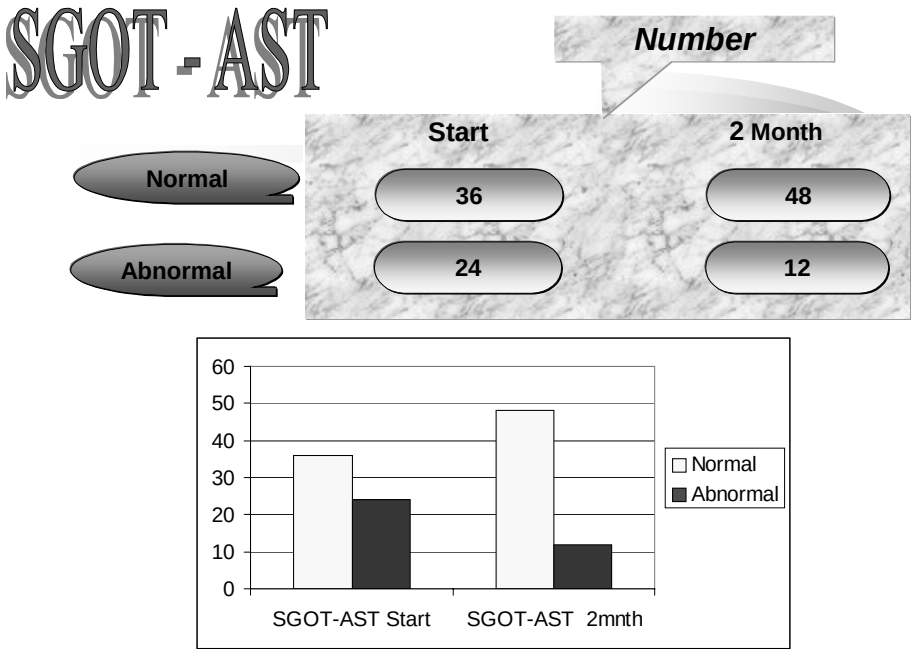
Number of PCR negative cases during and after ozone therapy

Fig. 4



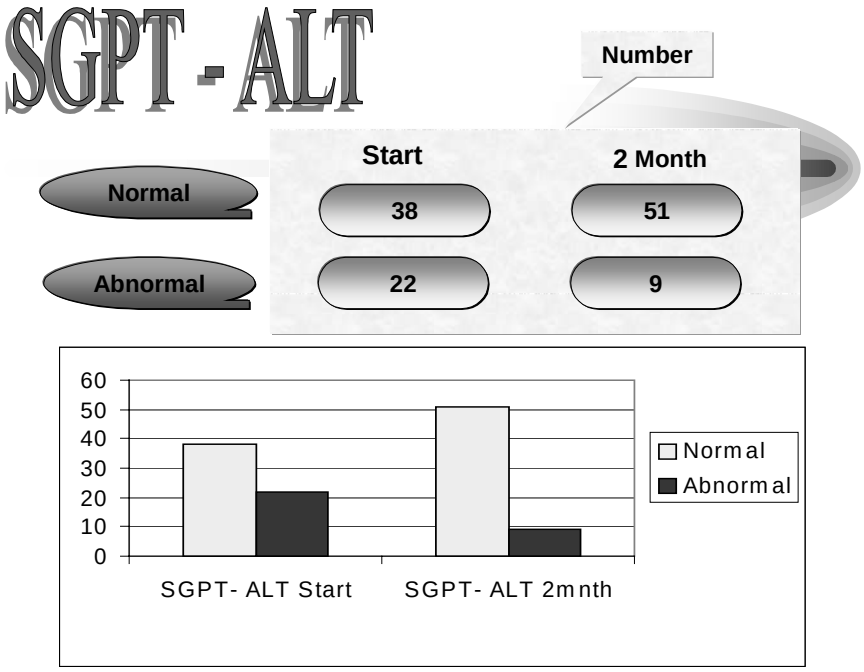
Number of cases with a decrease in PCR viral load during and after ozone therapy

Fig. 5



Normal and abnormal SGOT enzyme levels before and after ozone therapy

Fig. 6



Normal and abnormal SGPT enzyme levels before and after ozone therapy

Discussion

The significant decrease in viral load is an important factor – among other factors – for judging the improvement of a case of hepatitis C virus. In this study, it was found that following ozone therapy; there was a significant reduction of viral load. This decrease was evident after 8 weeks and further decline following another 16 weeks of ozone therapy.

Normal enzyme levels are a very important indicator denoting the sound integrity of liver cells. In this study, it was found that following ozone therapy; there was a significant change of abnormal enzyme levels towards normal values.

Some patients received DDB pills that are a Chinese herbal medicine capable of lowering the enzyme level, but without any anti-viral action. Stoppage of DDB will be followed by increase in enzyme level to the previous level. This can explain why some of the patients had a normal enzyme level before starting ozone therapy.

Clinical observations and questioning of the patients revealed that in 95 % of cases the general condition improved and some of patients returned to work after they were staying at home. Moreover in most cases there were improvement of the quality of life and they had the sense of well-being. All these data points to the important role of ozone a safe, effective method of therapy.

In 5% of cases the viral load did not decrease and were not responding to ozone therapy. Observation the patients and by clinical analysis reveled that there are three major factors for irresponsiveness: 1- improper diet with lots of fats and meat that is capable of lying great stress on the liver as an organ for metabolism and exhausting this organ. 2- physical exhaustion with lots of effort and long hours in work. This will through a great burden on the body as a whole and the liver as a part of the body. 3- hepatotoxic drugs that might be taken by the patient for treatment of another disease.

It is understandable that the response to treatment will be less in complicated cases with e.g. liver cirrhosis and ascitis or cases associated with chronic diseases e.g. diabetes and bilharziasis, but however, these were not considered as factors in ineffectiveness.

In this study there were no control group and the patient was considered a control to himself and the main issue, as a preliminary study was to compare the clinical and laboratory findings before and after ozone therapy.

In order to reach a proper protocol for ozone therapy, several pilot studies had to be accomplished. Trial MAH twice/week for 2 months, MAH three times/week for 2 months, RI twice/week for 2 months, RI three times /week for 2 months following rationale of start low and go slow. Good results were obtained but not as good as the study protocol.

After 8 weeks treatment as mentioned in this study protocol, combined MAH & RI once /week was tried as a pilot study for continuing therapy, but however the results less than in present study protocol from the general condition point of view. If we shift to twice / week the general condition is better.

Combination of MAH and RI was important the deliver ozone therapy to both systemic and portal circulation. Ozone therapy was found to induce hyper-oxygenation of portal circulation.

Conclusion

As a preliminary study Ozone therapy was found to be an effective, safe and less expensive method in Hepatitis "C" patients but further studies are important.

The protocol of ozone therapy in this study was found to be the best of many other protocols dealing with hepatitis C type 4.

Recommendations for further studies

It is recommended to start a double blind randomized placebo controlled Study. The patients must be selected with No complications (Cirrhosis, Ascitis, Liver cell failure, etc..) and No associated chronic disease (Diabetes, Bilharziasis, etc..) It is recommended to start a long-term study for one year and the follow-up by observation and investigations for another year. Evaluation should be based on many parameters i.e. general condition, liver Function tests (synthesis,excretions,integrity), quantitative PCR, abdominal ultrasonograpohy and liver biopsy. Quantitative PCR can be considered as a guide for evaluation but is not conclusive because of the sharp fluctuations of viral load and so far there is no precise and accurate method for quantitative estimation of viral load. Different methods, different units and wide variation from one laboratory to another must be put in consideration

References

- Bartenschlager R. Candidate targets for hepatitis C virus-specific antiviral therapy. Intervirology 1997; 40:378-393
- Bocci V, Luzzi E, Corradeschi F, Paulesu, et al. Studies on the biological effects of ozone: 5. Evaluation of immunological parameters and tolerability in normal volunteers receiving ambulatory autohaemotherapy. Biotherapy 1994; 7:83-90
- Bocci V. Ozonation of blood for the therapy of viral diseases and immunodeficiencies. A hypothesis. Medical Hypotheses 1992 Sept; 39(1):30-34
- Bolton DC, Zee YC, Osebold JW. The biological effects of ozone on representative members of five groups of animal viruses. Environmental Research 1982; 27:476-48
- Carpendale MT, Freeberg JK. Ozone inactivates HIV at noncytotoxic concentrations. Antiviral Research 1991; 16:281-292
- Dieperink E, Willenbring M, Ho SB. Neuropsychiatric symptoms associated with hepatitis and interferon alpha: A review. Am J Psychiatry 2000 June; 157(6):867-876
- Harrison TJ, Zuckerman AJ (Eds). The Molecular Medicine of Viral Hepatitis. Molecular Medical Science Series. John Wiley & Sons, New York, 1997
- Konrad H. Ozone therapy for viral diseases. In: Proceedings 10th Ozone World Congress 19-21 Mar 1991, Monaco. Zurich: International Ozone Association 1991:75-83
- Major ME, Feinstone SM. The molecular virology of hepatitis C. Hepatology 1997; 25: 1527-1538
- Maertens G, Stuyver L. Genotypes and genetic variation of hepatitis C virus. In: The Molecular Medicine of Viral Hepatitis. John Wiley $ Sons Ltd., London, 1997: 225-227
- Monjardino J. Molecular Biology of Hepatitis Viruses, Imperial College Press, London, 1998
- Paulesu L, Luzzi L, Bocci V. Studies on the biological effects of ozone: Induction of tumor necrosis factor (TNF-alpha) on human leucocytes. Lymphokine Cytokine Research 1991; 5:409-412
- Sunnen GV. Ozone in Medicine. Journal of Advancement in Medicine. 1988 Fall; 1(3): 159-174
- Viebahn R. The Use of Ozone in Medicine. Haug, Heildelberg, 1994
- Wells KH, Latino J, Gavalchin J, Poiesz BJ. Inactivation of human immunodeficiency virus Type 1 by ozone in vitro. Blood 1991 Oct; 78(7):1882-1890