

Syndrome of Malnutrition-Inflammation Complex in Chronic Alcoholics

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Abstract: Several studies have reported elevated serum ferritin levels in patients of chronic alcohol abuse. We hypothesized that serum ferritin may be acting as a marker of acute phase reactant, reflecting the inflammatory status of patients of chronic alcohol abuse, rather than as a marker of iron overload. Hence we correlated serum ferritin levels with interleukin 6 (IL-6), interleukin 1 (IL-1) and tumor necrosis factor alpha (TNF- α) which are markers of inflammation. Serum ferritin and IL-6 levels were significantly elevated at the time of admission. The elevation of IL-6 correlated well with the increase in ferritin levels ($r=0.78$, $p<0.01$). Serum iron levels were not significantly different before and after abstinence from alcohol. We also speculated that the inflammatory status was associated with the malnutrition that is observed in alcohol abuse. Therefore we studied serum total protein, albumin, prealbumin and transferrin levels as markers of nutrition in patients of chronic alcohol abuse. Serum levels of all these nutritional parameters increased significantly after 8 weeks of abstinence from alcohol and were accompanied by reduced levels of ferritin and IL-6. Hence it is possible that a malnutrition inflammation complex like syndrome exists in patients of alcohol abuse.

Key words: Ferritin, acute phase reactant, chronic alcohol abuse, Malnutrition – Inflammation – Complex Syndrome” (MICS)

INTRODUCTION: Serum ferritin is frequently reported to be elevated in chronic alcoholics.^{1,2} However these studies have indicated that the elevation of serum ferritin is a sign of iron overload.¹⁻⁴ Serum ferritin concentration results from the leakage of tissue ferritin, an intracellular iron storage protein. Serum ferritin is slightly different from tissue ferritin and contains little or no iron.⁵ While tissue ferritin clearly plays a role in intracellular iron handling, the role of serum ferritin is less clearly understood.⁶ Serum ferritin levels have been shown to increase during inflammation and the acute phase response.⁷ Inflammatory cytokines such as interleukin 1- β , (IL 1- β) and tumor necrosis factor alpha (TNF- α) increase the synthesis of ferritin.⁸ Chronic alcoholism is characterized by activation of the inflammatory response system and elevated serum levels of proinflammatory cytokines such as interleukin 6(IL-6) and tumor necrotic alpha (TNF- α).⁹⁻¹¹

Inflammation and protein energy malnutrition are closely related in hemodialysis patients.¹² The simultaneous combination of malnutrition and inflammation has been referred to as the “Malnutrition – Inflammation – Complex Syndrome” (MICS).¹³ This describes a syndrome whereby raised levels of pro-inflammatory cytokines (such as IL-1, IL-6 and TNF-alpha) are a common link between malnutrition, inflammation and atherosclerosis. MICS is believed to be the main cause of erythropoietin hypo responsiveness, high rate of cardiovascular atherosclerotic disease, decreased quality of life, and increased mortality and hospitalization in dialysis patients. Possible causes of MICS include co-morbid illnesses, oxidative and carbonyl stress, nutrient loss, anorexia and low nutrient intake, toxins, decreased clearance of inflammatory cytokines from the circulation.

Hence, we speculated that serum ferritin may be acting as an indicator of the acute phase reaction, reflecting the inflammatory status of patients of chronic alcohol abuse, rather than as a marker of iron overload. We correlated serum ferritin levels with interleukin 6 (IL-6), interleukin 1(IL-1) and tumor necrotic alpha (TNF - α) which are markers of inflammation. We also hypothesized that the inflammatory status was associated with the malnutrition that is observed in alcohol abuse. Therefore we studied serum total protein, albumin, prealbumin and transferrin levels as markers of nutrition in patients of chronic alcohol abuse.

We also compared the relationship of serum ferritin levels with serum gamma glutamyl transpeptidase (GGT). Serum GGT is known to be a marker of alcohol abuse. It is now also considered as a marker of oxidative stress and inflammation.¹⁴⁻¹⁶

This study was conducted among persons who had a long history of consuming alcohol. Patients were admitted into an alcohol rehabilitation unit and underwent abstinence from alcohol for a period of 8 weeks.

Our studies showed that serum ferritin levels correlated significantly with serum IL-6 and serum GGT levels in patients of chronic alcohol abuse. Levels of markers of nutrition like total protein and albumin which were reduced in patients of chronic alcohol abuse increased after 8 weeks of abstinence from alcohol and were accompanied by reduced levels of ferritin and IL-6. Hence it is possible that a malnutrition inflammation complex like syndrome exists in patients of alcohol abuse.

METHODS : Patients: Patients of chronic alcohol abuse (n=50) admitted to the “Deaddiction center for substance abuse, Kamineni Foundation”, Kamineni Institute of Medical Sciences, Narkatpally, Andhra Pradesh, India were inducted into the study. Demographic and clinical findings on admission. are shown in Table 1. All patients were males. Patients with existing liver disease and gross malnutrition were excluded from the study.

Table – I

Demographic and clinical findings on admission in 50 patients admitted to an alcohol withdrawal unit. All patients were males.

Age	:	39 \pm 12 years
Years of alcoholism	:	9 \pm 3
Nausea and vomiting	:	3
Chest pain, breathlessness, coughs	:	13
Headache, confusion, memory Loss	:	3
Burning in palms and feet	:	8
Abdominal pain	:	7
Generalized body Pains	:	7
Blurred vision	:	6
Tremors, Weakness in Limbs	:	9
Appetite Loss	:	10
Sleep disturbances	:	5
Ear pain / discharge	:	6

Pedal edema	:	3
Paralytic stroke	:	1
UTI	:	4
Diabetes	:	4
Hypertension	:	2

During admission, patients received a balanced nutritious diet and multivitamin tablets containing B₆, B₁₂ and folic acid. A qualified counselor provided psychological counseling for the patient and the patients' family. Other than treatment for minor ailments, patients did not receive any other medications during the 8-week period.

Written informed consent was obtained from each human subject and the procedures followed were in accordance with the ethical standards of the Institute Review Board of the Kamineni Institute of Medical Sciences and with the Helsinki Declaration of 1975, as revised in 1983.

Assays: Blood samples were obtained under fasting conditions on admission and after 8 weeks of alcohol abstinence. IL-1, IL-6 and TNF – alpha were measured by kits from Milenia Biotech GmbH (Germany). Serum bilirubin, total protein, albumin, SGPT, prealbumin and gamma glutamyl transpeptidase levels were measured on Cobas Integra 400 (Roche). Iron, TIBC, transferrin and ferritin were also measured by Cobas Integra 400 (Roche), The TIBC is calculated from the iron plus the unsaturated iron binding capacity.

The percentage transferrin saturation is calculated as serum iron / TIBC.

Statistical Analysis: For each parameter, mean and S.D. values were calculated. Comparisons between values before and after 8 weeks of alcohol abstinence were done by the t- test. A p value < 0.05 was required to reject the null hypothesis of no difference between the means. Pearson correlation was used to analyze the associations between variables.

RESULTS: A total of 50 subjects, all male were initially enrolled in the study. Of them, 8 patients dropped out of the program. Table – II shows the baseline mean and S.D. levels of iron indices, nutritional parameters, markers of inflammation and liver enzymes at the time of admission and following 8 weeks of abstinence from alcohol.

In general, the study patients were well nourished and did not show any signs of gross malnutrition, anemia or vitamin deficiencies. Their usual diet included rice, pulses, and green leafy vegetables with occasional dried fish and eggs. Even so all nutritional parameters (serum total protein, albumin, transferrin and prealbumin) increased significantly 8 weeks after abstinence from alcohol (Table-II). Serum ferritin and IL-6 levels were significantly elevated at the time of admission. The elevation of IL-6 correlated well with the increase in ferritin levels ($r=0.78$, $p<0.01$). Serum iron levels were not significantly different before and after abstinence from alcohol, indicating that iron overload in these patients was unlikely. The significant increase in iron binding capacity after abstinence from

alcohol is due to an, elevation of serum transferrin levels. This can be regarded as an index of improved hepatic protein synthesis.

Table II

Serum iron indices, parameters of nutrition and inflammation and liver enzymes were measured in patients of chronic alcohol abuse. Blood samples were drawn from the patients after an overnight fast. The first sample was taken within 2 days after admission and the second sample was taken following 8 weeks of abstinence from alcohol. Values are expressed as mean \pm S.D.

Parameter	Levels on admission (n=50)	Levels 8 weeks later (n = 42)	P value
Ferritin (ng/ml)	327.23 \pm 222.42	211.96 \pm 135.35	< 0.009
Transferrin (mg/dl)	263.58 \pm 72.19	303.67 \pm 83.50	< 0.01
Iron (μ g/dl)	118.22 \pm 66.42	114.98 \pm 55.05	N.S
TIBC (μ g/dl)	364.16 \pm 148.67	467.86 \pm 173	< 0.03
% saturation	34.92 \pm 20.05	29.25 \pm 15.8	< 0.08
GGT (IU/l)	43.72 \pm 37.03	25.9 \pm 8.97	< 0.03
SGPT (IU/l)	37.29 \pm 27.04	33.07 \pm 17.33	NS
Prealbumin (mg/dl)	29.14 \pm 11.35	25.76 \pm 8.50	< 0.05
Total protein (g/dl)	6.8 \pm 0.72	7.06 \pm 0.48	< 0.05
Albumin (g/dl)	3.8 \pm 0.57	4.02 \pm 0.40	< 0.02
IL-1 (pg/ml)	12.48 \pm 12.54	9.02 \pm 8.40	NS
IL-6 (pg/ml)	67.09 \pm 58.78	17.56 \pm 14.82	< 0.002
TNF – alpha (pg/ml)	20.73 \pm 31.77	16.86 \pm 33.7	NS

Serum GGT levels decreased significantly after abstinence from alcohol, signifying an improvement of liver function. Serum GGT levels correlated significantly with IL-6 (r=0.72,

p<0.05), with IL-1 (r=0.68, p<0.01) and with serum ferritin (r=0.62, p<0.05) at the time of admission. Serum GGT levels also showed a

negative correlation with serum total protein and albumin ($r = -0.45$ and -0.47 respectively).

The levels of SGPT and GGT at the time of admission in the patient were expressed as a ratio and patients were divided into 2 groups. Group I had an SGPT: GGT ratio of less than or equal to 1 while group II had an SGPT: GGT ratio greater than 1 (Table III). Patients with greater elevations of GGT signifying severe hepatic damage (group 1), had significant elevations of both, ferritin and IL-6. Following abstinence from alcohol for 8 weeks, GGT levels fell rapidly and all group I patients showed an increase in SGPT : GGT ratios.

Table – III:

Patients were classified into 2 groups based on the levels of GGT and SGPT at the time of admission. Patients with higher GGT elevations and having an SGPT: GGT ratio less than 1 were in Group – I and those with an SGPT: GGT ratio greater than 1 were in Group –II. Serum IL-6 and ferritin levels of both groups are given. (Mean + SD)

Parameter	Group – I (n=27)	Group – II (n=23)	p value
SGPT : GGT Ratio	0.61 ± 0.25	1.93± 0.94	< 0.03
Serum IL-6	63.27± 172.68	39.69± 21.02	<0.02
Serum ferritin	337± 172.68	220.88± 102.97	<0.008

Serum ferritin levels did not correlate significantly with serum albumin or serum total protein levels, indicating that ferritin itself did not have a direct effect on lowering the hepatic synthesis of proteins.

DISCUSSION: A derangement of the balance between proinflammatory and anti-inflammatory signals is characteristic of compensated and decompensated alcoholic cirrhosis.¹⁶ During the acute phase response of inflammation; inflammatory cytokines increase the synthesis of both H and L subunits of ferritin^{5,6}. In this study, patients of chronic alcohol abuse had significantly elevated IL-6 levels, which correlated with serum ferritin levels. This indicates that serum ferritin was associated with the proinflammatory cytokine and hence to the ongoing inflammatory process. Mean serum albumin and total protein were significantly reduced with chronic alcohol consumption. Albumin is considered to be a negative acute phase protein whose synthesis is suppressed by IL-6.¹⁷ Patients with chronic kidney disease with the malnutrition – inflammation complex syndrome (MICS) also presented with similar biochemical findings of elevated serum IL-6 and ferritin levels and lowered albumin and total protein.^{7,8}

In this study we have shown that 8 weeks of abstinence from alcohol reduced IL-6 and ferritin levels and increased albumin, total protein, prealbumin and transferrin levels. This agrees well with other studies where increased serum levels of IL-6, IL-10 and IL-8 have been reported to have declined few days after alcohol abstinence in patients with alcohol withdrawal syndrome.¹⁶ Mean serum iron

levels after abstinence were not significantly different from levels at the time of admission. Hence the possibility of excessive iron intake leading to elevated ferritin values is less likely.

In this study, serum ferritin levels were significantly elevated in the group of patients who had higher elevation of serum GGT at admission. This correlation between serum GGT and ferritin levels points to a connection between hepatic damage and inflammation. Therefore, in patients of chronic alcohol abuse, it would be more appropriate to regard elevated serum ferritin levels as an indicator of an inflammatory response and part of the malnutrition-inflammation complex syndrome.

ACKNOWLEDGEMENTS: The study was supported by a grant from the Kamineni Foundation. The technical assistance of Vinod Kumar is appreciated.

REFERENCES:

1. Worwood, M. (1990) Ferritin. *Blood Rev.* 4, 259-269
2. Yang SQ, Lin HZ, Yin M, Albrecht JH, Diehl AM. (1998) Effects of chronic ethanol consumption on cytokine regulation of liver regeneration *Am J Physiol.* 275 (4Pt 1):G696-704.
3. Whitfield, J. B. et al.(2001). Effects of alcohol consumption on indices of iron stores and of iron stores on alcohol intake markers. *Alcohol Clin Exp. Res.* 25, 1037-1045
4. Loannou, G.N. et al.(2004) The effect of alcohol consumption on the prevalence of iron overload, iron deficiency and iron deficiency anemia. *Gastroenterology* 126, 1293-1301.
5. Kalantar-Zadeh K, Don BR, Rodriguez RA, Humpherys MH (2001). Serum ferritin is a marker of morbidity and mortality in hemodialysis patients. *Am J Kidney Dis.* 37, 564-572.
6. Rogers JT. (1996) Ferritin translation by interleukin 1 and interleukin 6- the role of sequences upstream of the start codons of the heavy and light subunit genes. *Blood* 87, 2525-2537.
7. Rogers JT, Bridges KR, Durmowicz GP, Glass J, Auron PE, Munro HN. (1990). Translational control during the acute phase response. Ferritin response to interleukin 1. *J Biol Chem* 265:14572-14578.
8. Gonzalez-Quintela A. et al (2000) Influence of acute alcohol intake and alcohol withdrawal on circulating levels of IL-6, IL-8, IL-10 and IL-12. *Cytokine* 12, 1437 - 1440.
9. Lin ,H.Z.et al (1998) Chronic ethanol consumption induces the production of tumor necrosis factor-alpha and related cytokines in liver and adipose tissue.*Alcohol Clin Exp Res.* 22(5Suppl),231S-237S.
10. Khoruts, A. et al (1991) Circulating tumor necrosis factor, interleukin-1 and interleukin-6 concentrations in chronic alcoholic patients. *Hepatology.* 13, 267-76.

11. Kalantar – Zadeh, K, Rodriguez RA, Humpherys MH (2004) Association between serum ferritin and measures of inflammation, nutrition and iron in hemodialysis patients. *Nephrol Dial Transplant* 19, 141-149. patients. *Kidney international* 65, 1408–1415
12. Siren Sezer, F. et al (2002) Triad of Malnutrition, Inflammation, and atherosclerosis in hemodialysis patients. *Nephron* 91 , 456-462. 14
13. Alte, D. et al (2003) Distribution and dose response of laboratory markers to alcohol consumption in a general population; Results of the study of healthy in Pomerania (SHIP). *Journal of studies on Alcohol* 64, 75-82.
14. Chen, J. (2003) Combining carbohydrate – deficient transferrin and gamma glutamyl transferase to increase the diagnostic accuracy for problem drinking. *Alcohol and alcoholism* 38 , 574-582.
15. Singh, J. et al (1986) Gamma-Glutamyl transpeptidase: a novel biochemical marker in inflammation. *Biochem Pharmacol* 35 , 3753-60.
16. Daniluk J, Szuster-Ciesielska A, Drabko J, Kandefor-SzerszenM. (2001) Serum cytokine levels in alcohol-related liver cirrhosis. *Alcohol*. 23, 29-34
17. George A Kaysen, Joel A Dubin, Hans Georg Muller , (2004) Inflammation and reduced albumin synthesis associated with stable decline in serum albumin in hemodialysis