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Additive Value of Thyroid Uptake and Scintigraphy in Diagnosis of Short-Term Thyroid Dysfunction in Hepatitis C Patients Treated With Interferon-α

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ABSTRACT:

Objectives: The aim of this work was to assess the additive value of thyroid scan and uptake in the early diagnosis of interferon-α (IFN-α) induced thyroid dysfunction. **Methods:** Eighty HCV-positive Egyptian patients (30 males and 50 females, mean age 41.7±10.8 years) who received specific treatment in the form of IFN-α plus oral ribavirin. Patients with any thyroid disorder were excluded. Initial and 12 weeks after starting IFN-α treatment, all patients were subjected to 99mTc pertechnetate thyroid scan and uptake, free tetraiodothyronin (FT4), free triiodothyronin (FT3) and thyroid stimulating hormone (TSH), serum anti-thyroid autoantibodies including anti-thyroglobulin (TG-Ab), antiperoxidase (TPO-Ab) and anti-TSH (TSH-Ab). Neck US was done when indicated. **Results:** Seven patients (8.8%) developed hyperthyroidism (with elevated FT4, FT3 and suppressed TSH; mean 99mTc uptake = 10.5±5.15 vs 2.1±0.8% at base-line). Sixteen patients (20%) developed thyrotoxic phase of thyroiditis (with elevated FT4, FT3 and suppressed TSH; mean 99mTc uptake = 0.2±0.1 vs 1.0±0.6% at base-line). Significant elevation of TG-Ab and TSH-Ab was present in toxic patients compared with base-line. While, significant elevation of TG-Ab and TPO-Ab was present in thyroiditis patients compared with base-line. In toxic patients, 4/7 patients had clinical symptoms of thyrotoxicosis and US revealed hypervascularity in 3/7 patients while in thyroiditis patients, 8/16

patients had symptoms of transient thyrotoxicosis and US revealed features of destructive pattern in 5/16 patients. **Conclusions:** Thyroid dysfunction may occur as early as 3 months of starting IFN-α therapy in a significant number of HCV patients. Thyroid uptake and scan could differentiate between Graves' disease and transient thyrotoxic phase of thyroiditis allowing for accurate management and can be added as a part of routine investigation together with thyroid hormonal profile and anti-thyroid autoantibodies especially if other investigations are not diagnostic.

Keywords: HCV, Interferon-α, Thyroid dysfunction, auto thyroid antibodies, 99mTc pertechnetate thyroid uptake.

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INTRODUCTION:

The Middle East and North Africa region appears to have the highest prevalence of hepatitis C virus (HCV) worldwide [1, 2]. Egyptian population has one of the highest worldwide prevalence of HCV (15%) [3, 4 and 5]. IFN-α therapy alone or in combination with other drugs – is associated with many side effects ranging from Influenza-like symptoms to hematologic and neuropsychiatric side effects [6]. IFN-α has important immunomodulatory properties due to which it can induce autoimmune phenomena like autoimmune thyroiditis with hypo- or hyperthyroidism [7]. Autoimmune thyroiditis has been reported in up to 20% of the patients during IFN-based therapies [8]. Thyroid dysfunction may also manifest as destructive thyrotoxicosis, Graves' thyrotoxicosis and hypothyroidism. These pathological conditions may occur in the same patient as a result of different immunological effects of IFN-α therapy on the thyroid gland [9]. IFN treatment may also induce a subtle defect in the thyroidal organification of iodide, thus further impairing hormone synthesis [10]. One of the commonest side effects of IFN-α therapy is thyroiditis, with up to 40% of HCV patients on IFN-α developing clinical or subclinical disease. In some cases interferon induced thyroiditis (IIT) may result in severe symptomatology that may necessitate discontinuation of therapy [11, 12].

A common drug used with IFN-α in HCV treatment is Ribavirin (RIBA). RIBA is a synthetic analog of guanoside that induces the Th1 cytokines in the immune response against HCV infection [(13)].

When undergoing treatment, IFN and ribavirin synergize to stimulate the immune system in order to eradicate the virus that subsequently and indirectly affects the thyroid gland [(14)].

The correlation between the therapy and the gland malfunction that clinicians have often reduced the dose or sometimes even discontinued IFN-α treatment in patients who develop thyroid dysfunction, thus possibly compromising the therapeutic response [(15)].

In patients treated with IFN-α, the positivity of only TG-Abs at low titer is a specific marker of thyroid follicular destruction, whereas in the general population the presence of circulating TG-Abs is not considered an important risk factor for the development of thyroid dysfunction [(16, 17)].

IFN-α treatment may also induce Graves' hyperthyroidism [(18)]. The prevalence of this disorder is less frequent than other thyroid dysfunction because only 20–25% of all patients with IFN-α-related thyrotoxicosis are due to Graves' disease induced by circulating TSH-Abs [(19)].

The aim of this work was to assess the additive value of ^{99m}Tc pertechnetate thyroid scan and uptake in the early diagnosis of interferon-α (IFN-α) induced thyroid dysfunction in HCV patients.

PATIENTS and METHODS:

This prospective study was conducted on 80 HCV-positive Egyptian patients (grade 1, 2, 3 fibrosis). This study included 30 males (37.5%) and 50 females (62.5%), their mean age was 41.7±10.8 years, range (25-65 years). Patients were eligible to receive specific treatment for HCV according to the standard institutional protocol in the form of IFN-α plus oral ribavirin. IFN-α was given as subcutaneous injection once per week for 48 weeks. Routine laboratory investigations were performed before starting treatment course including Liver enzymes (ALT&AST), serum albumin, bilirubin, I.N.R., random blood sugar, renal function tests, fundus examination and E.C.G. exclusion criteria of the current study included patients with history of thyroid disease, any associated other autoimmune diseases (e.g. of rheumatic cause), patients with recent history of CT with contrast and patients on medications that could interfere with ^{99m}Tc pertechnetate uptake (e.g. amiodarone).

Patients with base line abnormalities in the thyroid uptake level, thyroid ultrasonography or thyroid hormonal profile were also excluded. The study was approved by the institutional ethical committee. **Methods:** At base line and 12 weeks after starting IFN-α treatment, all patients were subjected to ^{99m}Tc pertechnetate thyroid scan and uptake, thyroid hormones in the serum including FT3, FT4 and TSH using radioimmunoassay, serum anti-thyroid autoantibodies including anti-thyroglobulin (TG-Ab), anti-peroxidase (TPO-Ab) and anti-TSH (TSH-Ab) using ELIZA lab technology. Table 1 displays the normal ranges of the thyroid hormones and autoantibodies. Neck ultrasonography was done when indicated.

Table 1: Normal values of thyroid hormones and anti-thyroid antibodies.

Test	Normal values	Interpretation
Free T3	2-4.4 pg/ml	
Free T4	0.9-1.7 ng/dl	
TSH	0.27-4.2 uIU/ml	
Anti-Thyroglobulin Antibodies	< 0.6 IU/ml 0.6-1 IU/ml > 1 IU/ml	-ve Weak +ve Moderate to strong +ve
Anti-Peroxidase Antibodies	< 50 IU/ml 50-75 IU/ml > 75 IU/ml	-ve Borderline High
Anti-TSH Antibodies	< 30 IU/ml	

^{99m} Tc pertechnetate thyroid scan imaging protocol: No special patient's preparation was needed. ^{99m}Tc pertechnetate was injected in a dose of 185 MBq (5 mCi) through intravenous followed by saline wash. Patients positioned supine under the gamma camera (dual head, Siemens, Symbia connected to Dell processing unit) mounted with a low energy, parallel-hole collimator, 15% energy window centered at 140 KeV, 256x256 matrix for static images and zoom 2.23. Count of the syringes before and after injection for 1 minute was obtained. Anterior static images of the neck for 500,000 counts, 20 minutes post-injection were acquired with the neck extended to the closest distance from the collimator. Thyroid uptake values were estimated automatically using the pre and post syringe calculated method (normal range of uptake values is 0.5-4 % in our institution).

Statistical Analysis: Data were entered into the Statistical Package of Social Science Software program version 21 (SPSS) to be statistically analyzed. Data were summarized using mean, standard deviation and median for quantitative variables, and frequency and percentage for qualitative ones. Comparison between groups was performed using unpaired student's t-test or Mann–Whitney-Wilcoxon test for quantitative variables and Chi square or Fisher's exact test for qualitative ones. P values less than 0.05 were considered statistically significant. Graphs were used to illustrate some information.

RESULTS:

According to the results of thyroid hormones and scan findings on follow up, 23/80 patients (28.7%) developed thyroid dysfunction in the form of thyrotoxicosis or thyroiditis. Thyrotoxicosis was developed in 7 patients (8.8%), while 16 patients (20%), developed thyroiditis. The remaining 57 patients (71.3%) showed normal serum FT3, FT4 and TSH levels and normal scan pattern and uptake (Table 2). Thyroid hormonal profile at base line and 3 months after treatment is seen in ***table 2***. No changes could be elicited in the normal group of patients. While in toxic and thyroiditis patients, a statistically significant change in the serum level of FT3, FT4 and TSH were found **($p < 0.001$)**. The rise in serum FT3 and FT4 was significantly higher in toxic patients more than patients with thyroiditis **($p < 0.001$)**. While TSH suppression between both groups was statistically non-significant **($p = 0.999$).**

Table 2: Thyroid hormonal profile and ^{99m}Tc thyroid uptake % at base line and 3 months after IF-α treatment in 80 patients with HCV.

		Normal (N=57) (mean±SD)	Toxic (N=7) (mean±SD)	Thyroiditis (N=16) (mean±SD)
FT3 (pg/ml)	**Baseline**	2.8±0.6	2.8±0.6	2.6±0.8
	After 3m	3.2±1.2	9.1±4.0	5.3±0.5
FT4 (ng/dl)	**Baseline**	1.2±0.2	1.2±0.3	1.3±0.2
	After 3m	1.3±0.3	5.9±4.0	3.2±1.2
TSH* (uIU/ml)	**Baseline**	1.6±1.1	1.7±1.0	1.6±1.2
	After 3m	2.3±1.1	0.07±0.04	0.08±0.05
Thyroid uptake %	**Baseline**	1.9±0.9	2.1±0.8	1.0±0.6
	After 3m	2.1±1.1	10.5±5.1	0.2±0.1

The percentage of thyroid uptake and thyroid scan results after 3 months could accurately differentiate between thyroid gland toxicity **(fig. 1)** and transient thyrotoxicosis of thyroiditis **(fig. 2)** (mean uptake value = 10.5±5.1% vs 0.2±0.1%, respectively; **(P < 0.001).**

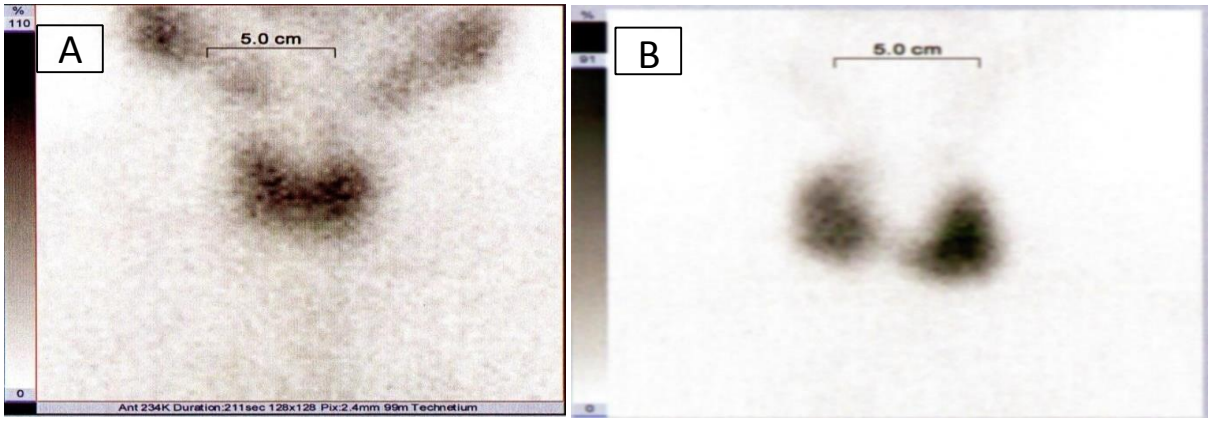

Fig. 1 Female patient, 44 years old with HCV under interferon therapy developed symptoms of thyrotoxicosis. **A**. At base line, normal thyroid scan and uptake level = 2.5%. **B.** Follow up after 12 weeks, thyroid scan showed features of Graves' disease with intense uniform tracer distribution and high uptake level = 11.2%.

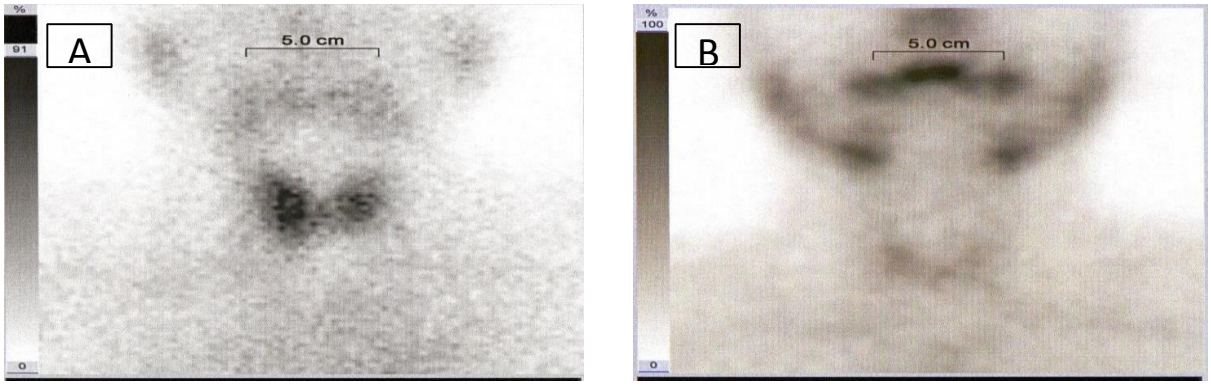

Fig. 2 Male patient, 51 years with HCV under interferon therapy developed thyroiditis. A. At base line, normal thyroid scan and uptake level = 0.7%. B. Follow up after 12 weeks, thyroid scan showed features of thyroiditis with faint tracer distribution and low uptake level = 0.1.

Anti-thyroid Antibodies results: ***Table 3*** demonstrates the frequency of patients with elevated anti-thyroid Abs at basal condition and 3 months after starting treatment. Twenty nine patients (36%) had base line elevation of anti-thyroid autoantibodies. Do novo elevation of anti-thyroid Abs was found in 15 patients (18.7%), being 44 patients (55%) after 3 months. A statistically significant difference between groups was elicited only with anti TG-Ab ($p<0.01$).

Anti TG-Ab: A higher mean base line serum anti TG-Ab was found in thyroiditis group (mean = 1.0±0.9 IU/ml) compared with that in normal and toxic groups (mean = 0.4±0.6 and 0.3±0.2 IU/ml, respectively) ($p = 0.008$). The rise in anti TG-Ab after 3 months was statistically significant in thyroiditis and toxic groups (mean = 2.3±1.2 and 1.5±1.3 IU/ml, respectively) compared with the normal group (mean = 0.6±0.5 IU/ml) ($p = 0.001$) ***(Table 3 and fig. 3A)***. **Anti-peroxidase Ab:** Mean basal serum anti-peroxidase Ab was within normal range in all groups. The rise was elicited after 3 months in thyroiditis group only (mean = 97.9±75.0 IU/ml) compared with the normal and toxic groups (mean = 41.2±21.2 and 38.7±7.0 IU/ml, respectively) ($p = 0.001$) (Table 3 and fig. 3B). **Anti-TSH Ab:** Mean basal serum anti-TSH Ab was within normal range in the normal, toxic and thyroiditis groups (13.2±9.0, 26.4±11.1 and 14.5±6.0, respectively).

The rise was elicited after 3 months in toxic group only (mean = 47.4±28.0 IU/ml) compared with the normal and thyroiditis groups (mean = 20.3±8.8 and 19.5±9.0 IU/ml, respectively) ($p = 0.036$) (Table 3 and fig. 3C). Pearson correlation analysis between thyroid uptake % and anti-thyroid antibodies after 3 months among all groups showed non-significant correlation.

Table 3: Mean values of anti-thyroid antibodies at basal condition and 3 months after treatment.

Anti-Thyroid Ab	Groups							
	Normal N=57		Toxic N=7		Thyroiditis N=16		Chi-Square	
	Base-line mean±SD	After 3m mean±SD	Base-line mean±SD	After 3m mean±SD	Base-line mean±SD	After 3m mean±SD	X^2	p-value
Anti TG-Ab IU/ml	0.4±0.6	0.6±0.5	0.3±0.2	1.5±1.3*	1.0±0.9**	2.3±1.2*	8.145	0.017**
Anti-peroxidase Ab IU/ml	25.4±17.1	41.2±21.2	21.1±7.7	38.7±7.0	41.1±23.0	97.9±75.0*	2.081	0.353
Anti TSH Ab IU/ml	13.2±9.0	20.3±8.8	26.4±11.1	47.4±28.0*	14.5±6.0	19.5±9.0	3.571	0.168

* Mean values are higher than normal reference. ** P value is significant if < 0.05

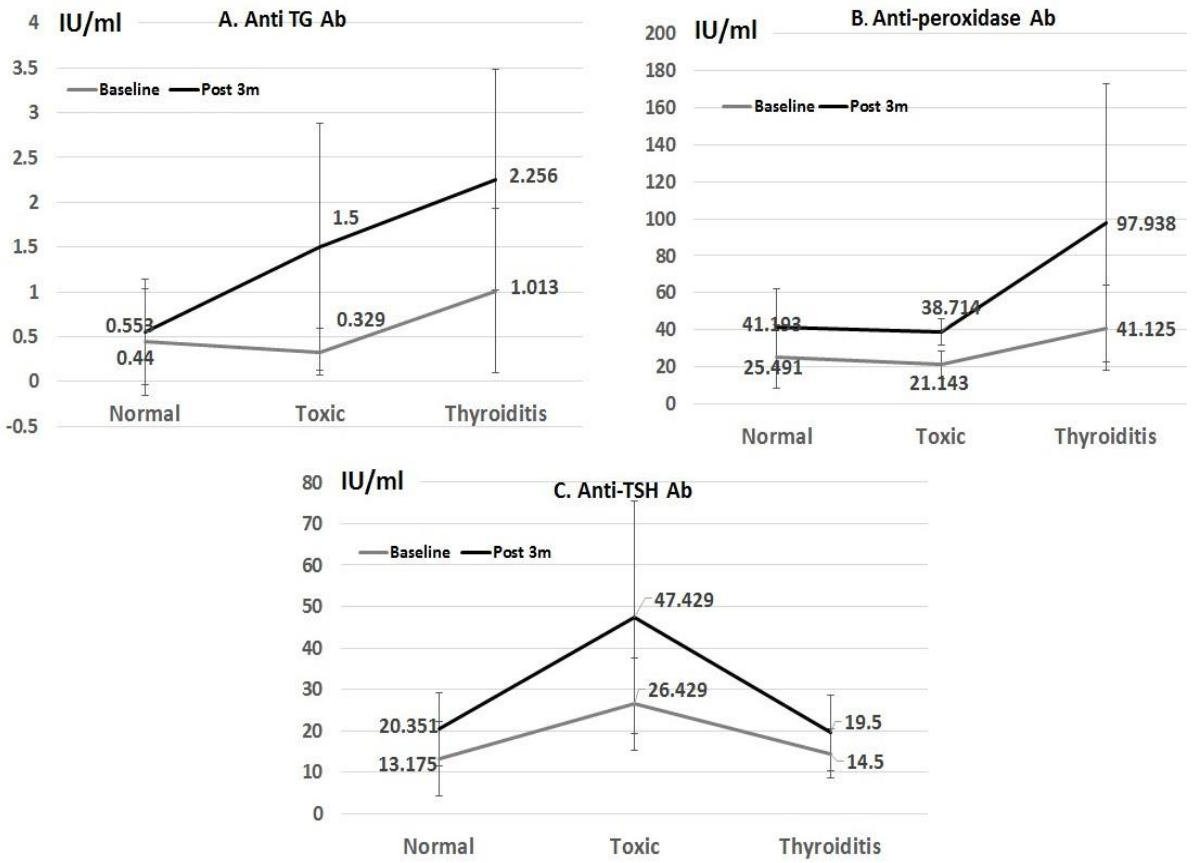

Fig. 3 (A, B, C): Mean values of the serum levels of auto thyroid antibodies in 80 patients with HCV before and 12 weeks after IF-α treatment. **A.** Anti-thyroglobulin Ab, **B**. Anti-peroxidase Ab **C.** Anti-TSH Ab.

Baseline US was done in 35 patients who had suspicious clinical neck lesions to exclude underlying thyroid pathology and revealed normal thyroid morphology. Ten patients were excluded from the study after US examination due to the presence of thyroid nodules despite their normal thyroid hormonal profile. At follow up after 3 months, neck US was performed to patients in the toxic and thyroiditis groups. In toxic group (N=7), US revealed hypervascularity of the thyroid gland in 3 patients (42.8%) while in thyroiditis group (N=16), US revealed features of hypo echogenicity, hypo vascularity and destructive pattern in 5 patients (31.3%). At time of follow up after 3 months of initiation of IF-α treatment, symptoms of hyperthyroid state (mainly palpitation) were documented by 4 patients (57.1%) in toxic group who required anti-thyroid medication. In thyroiditis group, 8 patients (50%) had thyrotoxic symptoms and no need for specific treatment.

DISCUSSION:

In HCV patients treated with IF-α, activation of the immune system is important for the development of thyroid disease. Furthermore, IF-α has direct inhibitory effects on thyroid hormone synthesis, release, and metabolism [(20)].

A genetic predisposition to thyroid autoimmune disease is probably necessary for the development of thyroid disease in patients treated with IF-α [(8, 21)]. However, the wide spread effects of IF-α on the immune system may be important for inducing thyroid disease because peripheral features of systemic immune involvement have been described in patients with thyroid autoimmunity [(22,23)]. In addition to the systemic effects, IF-α may have direct effects on the thyroid gland by modulating the aberrant expression of major histocompatibility antigens on thyroid cells [(24)] and favoring a cytokine microenvironment, which may lead to the immune-mediated damage of thyroid tissue [(25)]. The study aimed at verifying the possible influence of IFN-α therapy in inducing clinical, laboratory and scintigraphic signs of altered thyroid function. In the current study, there was no evidence of clinical signs of systemic autoimmune disease in HCV patients positive for non-organ-specific autoantibodies in basal condition before IF-α therapy. The short follow-up period allowed to verify the occurrence of early thyroid gland dysfunction after 3 months related to treatment with IF-α, with 28.7% of patients developed thyroid dysfunction based on the results of thyroid hormones,

Thyroid scan and uptake values. This is in concordance with ***Rocco et al.,*** [26] who reported a 27.3% prevalence of thyroid dysfunctions in their patients. Other studies [27, 28] described less frequent thyroid dysfunction in 15% of all treated patients. The higher percentage in our results could be related to the higher number of females patients (50/80, 62.5%) who show a higher natural incidence of autoimmune reactions if compared to male population as reported in other studies [29, 30]. The age of patients seems of negligible importance in this study with mean age of patients in the 3 groups is almost similar. However, the prevalence of thyroid autoantibodies in ordinary general population usually increases with age [31]. Since our HCV patients were selected on the basis of absence of thyroid abnormality, the significant elevation of thyroid autoantibodies after therapy seems to support the postulation that the autoimmune reactions are likely due to the immunomodulatory properties of IF-α rather than to the exacerbation of preexisting thyroid abnormalities [26]. The present study revealed positive influences of IF-α therapy on the thyroid autoantibodies with statistically significant elevation of thyroid autoantibodies was detected in 44 patients compared to 29 patients at basal condition which reflects possible do novo thyroid autoimmunity in 15 patients (18.7%) similar to that reported by other studies [26,32]. Destructive thyrotoxicosis usually occurs in the first weeks of IF-α treatment in close temporal relationship with the appearance of thyroid autoantibodies, especially anti thyroglobulin antibodies [16]. In the present study, destructive thyroiditis was confirmed in 16 patients (20%) with elevated anti TG-Ab and negative anti TSH-Ab [33] in all of them. We found that, only half of patients with destructive thyroiditis had transient manifestations of thyrotoxicosis. In addition to the thyroid hormonal profile, thyroid scan pattern with low uptake values could accurately diagnose thyroiditis in the remaining 50% who had mild and transient thyrotoxicosis without overt clinical manifestations. Furthermore, the introduction of thyroid scan and uptake in the current study; at time of expected thyroid dysfunction shortly after IFN-α therapy; could definitely differentiate between either thyrotoxicosis due to excessive release of thyroid hormones by the damaged thyroid follicles in thyroiditis group or thyrotoxicosis due to Graves' disease in toxic group of patients. Thyroid ultrasonography, thyroid hormones and

clinical symptoms cannot be diagnostic in such condition. ***Wong et al*** [18] reviewed the results of their HCV patients who were treated with IFNα over a period from 1996 to 2001 and reported only 10 patients developed thyrotoxicosis. In our prospective study, we reported an incidence of 7 patients with positive clinical symptoms , 4 of them also had high anti TSH-Abs, such finding supposes that, the true prevalence of this disorder is much higher than that reported in the literature because it is often transient and has mild clinical manifestations [16,18]. In the present study, anti-peroxidase Ab was elevated in about half of patients who developed thyroid dysfunction after therapy in concomitant with other autoantibodies (anti TG-Ab and anti TSH Ab). Although it is uncertain whether antiperoxidase Ab has a major pathophysiological role in inducing hypothyroidism [33], it has been suggested that these antibodies reflect a more advanced and aggressive underlying autoimmune destructive process of the thyroid gland [21,34]. We recommend screening all hepatitis C patients for thyroid disease prior to starting IF-α therapy. Baseline screening should include at least TSH levels but we recommend screening also for thyroid autoantibodies since positive Abs are a significant risk factor for IFN-α to initiate thyroid dysfunction and should be monitored closely to avoid complications of thyroid disease such as cardiac arrhythmias. Also, if the patient develops abnormal thyroid functions while on IFN-α, a full workup including serum TSH, thyroid autoantibodies as well as thyroid uptake and scan to distinguish between Graves' disease and destructive thyroiditis.

CONCLUSIONS:

Thyroid dysfunction may occur as early as 3 months of starting IFN-α therapy in a significant number of HCV patients. Thyroid uptake and scan could differentiate between Graves' disease and transient thyrotoxic phase of thyroiditis allowing for accurate management and can be added as a part of routine investigation together with thyroid hormonal profile and anti-thyroid autoantibodies especially if other investigations are not diagnostic.

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