

LETTERS TO THE EDITOR

SIMILARITIES OF IMMUNE REACTIONS BETWEEN HEPATITIS C AND SEVERE ACUTE RESPIRATORY SYNDROME-ASSOCIATED CORONAVIRUS INFECTIONS

To the Editor,

Severe acute respiratory syndrome (SARS) is an emerging disease caused by a novel coronavirus (SARS-CoV). It has caused worldwide outbreaks, resulting in 8102 cases in 29 countries, with 744 fatalities in 2003.¹ SARS-CoV belongs to the *Coronaviridae* genus, and hepatitis C virus (HCV) belongs to the *Flaviviridae* genus. These are two different viruses, but both are positive single strain RNA viruses (+ssRNA), which have some similar characteristics in immune responses that might offer some insight into the treatment of SARS.

Cinatl *et al.* first reported that interferon beta could inhibit SARS-CoV replication in *in vitro* culture.² A similar result was confirmed by Hensley *et al.*³ Furthermore, other forms of interferon (interferon-alpha and -gamma) have also had an anti-SARS effect in *in vitro* studies.^{2,4} Loutfy *et al.* reported a potential benefit from the interferon alfacon-1 plus corticosteroids in SARS treatment, and found that the treatment group seemed to have reduced disease-associated impaired oxygen saturation, more rapid resolution of radiographic lung abnormalities and lower levels of creatine kinase.⁵ Zhao *et al.* also reported that SARS patients randomly receiving interferon alpha tended to have the shortest admission days, time needed for improvement of dyspnea and time for improvement of chest X-ray films than other treatment regimens.⁶ Ribavirin alone was not efficacious in *in vitro* anti-SARS-CoV test, or in the Zhao's randomized trial,⁶ although some have suggested its clinical value.⁷ Cinatl and colleagues⁸ tested certain potential anti-SARS-CoV drugs including ribavirin, 6-azauridine, pyrazofurin, mycophenolic acid and glycyrrhizin, and showed that glycyrrhizin has the best potency in inhibiting the replication of SARS-CoV. These results suggest that interferon alpha and glycyrrhizin could be beneficial in SARS treatment. Similarly, interferon alpha and glycyrrhizin are also effective in treating HCV infections.^{9,10} Glycyrrhizin lowers serum ALT during treatment, reduces the inflammation of HCV infection, and prevents the development of hepatoma.^{9,10} Glycyrrhizin has fewer side-effects and seems more effective than ribavirin in treating HCV infections. Taken together, these results suggest that SARS-CoV and HCV might have similar immunopathogenesis.

Patients with chronic HCV infection tend to have Th2 dominance.¹¹ In a previous study,¹² we also found, as did Wang *et al.*,¹³ that a Th2 predominant cytokine

profile with increases of TGF β and IL-13 in the early stages of SARS infection (within 7 days after symptoms), followed by a Th1 predominant cytokine profile with elevation of IL-2 and IL-18 in the late stage (≥ 2 weeks after symptoms). This suggests that the host immune response in early SARS infection might favor viral replication, but 2–3 weeks after SARS-CoV infection, the host immune response can induce specific immunity for virus clearance or immune-mediated damage. Based on the skewed immune responses, we therefore postulate to use interferon alpha in the early stage of SARS infection to deviate Th2 predominant reaction to Th1 reaction, and hold back corticosteroids until appearance of late exacerbation showing augmented Th1 response.

Another similarity between HCV and SARS-CoV infections is related to elevation of blood IL-8 levels. Polyak *et al.* have reported that chronic HCV infection with elevated IL-8 levels is associated with the resistance to interferon therapy.^{14,15} One possible mechanism might be related to activation of p38 MAP kinase that is associated with elevated IL-8 levels and resistant interferon therapy.¹⁶ We have recently shown that SARS infections were associated with mild elevation of IL-8 in conjunction with p38 activation in CD14⁺ cells.¹² How to prove the relationships between p38 activation-related interferon resistance and SARS-CoV- or HCV-related immune response deserves further investigation.

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REFERENCES

- 1 Severe Acute Respiratory Syndrome (SARS)—Frequently Asked Questions about SARS. Available from: <http://www.ndsc.ie/DiseaseTopicsA-Z/SevereAcuteRespiratorySyndrome/d689.PDF> pp. 1–4 (April 26, 2004).
- 2 Cinatl J, Morgenstern B, Bauer G, Chandra P, Rabenau H, Doerr HW. Treatment of SARS with human interferons. *Lancet* 2003; **362**: 293–4.
- 3 Hensley LE, Fritz LE, Jahrling PB, Karp CL, Huggins JW, Geisbert TW. Interferon-beta 1a and SARS coronavirus replication. *Emerg. Infect. Dis.* 2004; **10**: 317–19.
- 4 Stroher U, DiCaro A, Li Y *et al.* Severe acute respiratory syndrome-related coronavirus is inhibited by interferon-alpha. *J. Infect. Dis.* 2004; **189**: 1164–7.
- 5 Loutfy MR, Blatt LM, Siminovitch KA *et al.* Interferon alfacon-1 plus corticosteroids in severe acute respiratory syndrome: a preliminary study. *JAMA* 2003; **290**: 3222–8.
- 6 Zhao Z, Zhang F, Xu M *et al.* Description and clinical treatment of an early outbreak of severe acute respiratory

- syndrome (SARS) in Guangzhou, PR China. *J. Med. Microbiol.* 2003; **52**: 715–20.
- 7 Mazzulli T, Kain K, Butany J. Severe acute respiratory syndrome: overview with an emphasis on the Toronto experience. *Arch. Pathol. Lab. Med.* 2004; **128**: 1346–50.
 - 8 Cinatl J, Morgenstern B, Bauer G, Chandra P, Rabenau H, Doerr HW. Glycyrrhizin, an active component of liquorice roots, and replication of SARS-associated coronavirus. *Lancet* 2003; **361**: 2045–6.
 - 9 van Rossum TG, Vulto AG, Hop WC, Brouwer JT, Niesters HG, Schalm SW. Intravenous glycyrrhizin for the treatment of chronic hepatitis C: a double-blind, randomized, placebo-controlled phase I/II trial. *J. Gastroenterol. Hepatol.* 1999; **14**: 1093–9.
 - 10 Kumada H. Long-term treatment of chronic hepatitis C with glycyrrhizin (stronger neo-minophagen C [SNMC]) for preventing liver cirrhosis and hepatocellular carcinoma. *Oncology* 2002; **62** (Suppl. 1): 94–100.
 - 11 Kobayashi K, Ishii M, Igarashi T *et al.* Profiles of cytokines produced by CD4-positive T lymphocytes stimulated by anti-CD3 antibody in patients with chronic hepatitis C. *J. Gastroenterol.* 1998; **33**: 500–7.
 - 12 Lee CH, Chen RF, Liu JW *et al.* Altered p38 mitogen-activated protein kinase expression in different leukocytes with increment of immunosuppressive mediators in patients with severe acute respiratory syndrome (SARS). *J. Immunol.* 2004; **172**: 7841–7.
 - 13 Wong CK, Lam CW, Wu AK *et al.* Plasma inflammatory cytokines and chemokines in severe acute respiratory syndrome. *Clin. Exp. Immunol.* 2004; **136**: 95–103.
 - 14 Polyak SJ, Khabar KS, Rezeiq M, Gretch DR. Elevated levels of interleukin-8 in serum are associated with hepatitis C virus infection and resistance to interferon therapy. *J. Virol.* 2001; **75**: 6209–11.
 - 15 Polyak SJ, Khabar KS, Paschal DM *et al.* Hepatitis C virus nonstructural 5A protein induces interleukin-8, leading to partial inhibition of the interferon-induced antiviral response. *J. Virol.* 2001; **75**: 6095–106.
 - 16 Balasubramanian A, Ganju RK, Groopman JE. Hepatitis C virus and HIV envelope proteins collaboratively mediate interleukin-8 secretion through activation of p38 MAP kinase and SHP2 in hepatocytes. *J. Biol. Chem.* 2003; **278**: 35755–66.

LACK OF PBC-SPECIFIC ANTIMITOCHONDRIAL ANTIBODIES IN PATIENTS WITH *CHLAMYDIA PNEUMONIAE* INFECTION

To the Editor,

We searched for antimitochondrial antibodies (AMA) in sera from women with serologically-proven *Chlamydia pneumoniae* infection and matched controls to indirectly confirm the recently suggested role for the bacterium in the induction of primary biliary cirrhosis (PBC). Using the most sensitive methodology for AMA determination, none of the investigated sera were found positive for AMA.

The etiopathogenesis of primary biliary cirrhosis (PBC), an immune-mediated biliary disease, is still

unknown.¹ Autoantibodies directed against AMA can be detected in 95% of patients with PBC and appear to be highly specific for the disease. The cloning, expression and identification of major B-cell epitopes have greatly facilitated the specific and sensitive detection of AMA.²

It has been proposed that infectious agents or other environmental factors (bacteria, viruses or xenobiotics) might contribute to the breakdown of tolerance, leading to autoimmunity in PBC.¹ Molecular mimicry by specific exogenous proteins, which might induce cross-reactive autoimmune responses to host proteins, might be involved. Interestingly, Abdulkarim *et al.* have recently found *C. pneumoniae* antigens in 25/25 (100%) of liver samples from patients with PBC but only in 8.5% of controls, thus suggesting a role for this bacterium in the pathogenesis of PBC.³ However, these data could not be confirmed by others by using a sensitive polymerase chain reaction (PCR)-based method.⁴

Based on the hypothesis that *C. pneumoniae* might be a major external trigger in the pathogenesis of PBC, we sought AMA in a series of women with well-defined chronic *C. pneumoniae* infection. They were diagnosed by both positive nested touch-down PCR on peripheral blood mononuclear cells and positive microimmunofluorescence antibody testing (IgG ≥ 1:64 or IgG > 1:64 + IgA > 1:16).⁵ We note that *C. pneumoniae* seroprevalence in the middle-aged general population is known to be 55–60%.⁵

Sera from 48 women with *C. pneumoniae* chronic infection (mean age ± SD: 58.3 ± 7.3) and 56 age-matched healthy women (mean age ± SD: 58.7 ± 8.3) were tested for AMA. All subjects were of Italian descent and presented normal values of serum alkaline phosphatase at the time of enrollment. The latter feature was ascertained in order to attempt the identification of PBC cases at early stages. Women with *C. pneumoniae* infection were positive for active infection at PCR and presented titers of specific IgG, IgM, and IgA of 164 ± 231, 25 ± 39, and 5 ± 22, respectively.

We sought antibodies against the three major PBC-specific mitochondrial epitopes by using ELISA with the triple-expression hybrid clone, pML-MIT-3, as previously described.² Briefly, the AMA-reactive immunodominant epitopes within the three distinct lipoyl domains of human pyruvate dehydrogenase complex (PDC-E2), bovine branched-chain alpha-ketoacid dehydrogenase (BCOADC-E2) and rat oxoglutarate dehydrogenase complex (OGDC-E2) were cloned and coexpressed in the plasmid vector, pGEX-4T-1 (Pharmacia, Alameda, CA, USA), and were used as the antigen.

None of the investigated sera from both the patients with *C. pneumoniae* infection and the controls were positive for AMA. Accordingly, by using an indirect approach to assess the role of *C. pneumoniae* in PBC, we were unable to support the involvement of this bacterium in the induction of the disease. A possible reason for the negative findings could be that the number of patients with *C. pneumoniae* studied might have been too small. We also suggest that the analysis of a large number of PBC sera with different AMA patterns for signs of *C. pneumoniae* infection should be pursued.

However, based on the above considerations and data, we believe that the etiopathogenesis of PBC remains unknown and the search for responsible genetic or environmental factors continues.

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REFERENCES

- 1 Talwalkar JA, Lindor KD. Primary biliary cirrhosis. *Lancet* 2003; **362**: 53–61.
- 2 Moteki S, Leung PS, Coppel RL *et al*. Use of a designer tripe expression hybrid clone for three different lipoyl domain for the detection of antimitochondrial autoantibodies. *Hepatology* 1996; **24**: 97–103.
- 3 Abdulkarim AS, Petrovic LM, Kim WR, Angulo P, Lloyd RV, Lindor KD. Primary biliary cirrhosis: an infectious disease caused by *Chlamydia pneumoniae*? *J. Hepatol.* 2004; **40**: 380–84.
- 4 Leung PS, Park O, Matsumura S, Ansari AA, Coppel RL, Gershwin ME. Is there a relation between Chlamydia infection and primary biliary cirrhosis? *Clin. Dev. Immunol.* 2003; **10**: 227–33.
- 5 Blasi F, Cosentini R, Schoeller MC, Lupo A, Allegra L. Chlamydia pneumoniae seroprevalence in immunocompetent and immunocompromised populations in Milan. *Thorax* 1993; **48**: 1261–3.

TWO MALE PATIENTS WITH WILSON'S DISEASE TREATED USING TRIENTINE AND IRON REDUCTION THERAPY

To the Editor,

Ceruloplasmin plays an important role in iron transport because it is a major ferroxidase,¹ and genetic defects involving ceruloplasmin synthesis cause severe iron overload in the liver and brain.² The introduction of penicillamine³ has provided significant advances in the treatment of Wilson's disease and now trientine,⁴ a safer but less potent copper chelating agent, is available to patients who are intolerant to penicillamine. Penicillamine or trientine administration might reduce a patient's level of internally bioavailable copper, thereby leading to impaired iron transport. In a previous study, it was found that penicillamine further reduces serum ceruloplasmin levels and causes iron to accumulate in the liver.⁵

In the present letter, we report on two male patients with Wilson's disease who were treated using trientine and phlebotomy. One of the patients developed myasthenia gravis at the age of 30 years. He was compound heterozygous for 453delC and 2871delC in the *ATP7B*

gene, but negative for C282Y and H63D in the *HFE* gene. Liver histology showed chronic hepatitis with histochemical iron deposits, and his serum ferritin concentration was 277 ng/mL. Also, his serum ceruloplasmin and ferroxidase levels were low at 2 mg/dL and 240 U/L, respectively. One month after discontinuing penicillamine treatment, trientine hydrochloride was initiated at 2500 mg/day. The other patient was a 37-year-old businessman who presented with an intentional hand tremor. The results of liver histology, where some regenerative nodules were positive for copper and/or iron grains, were suggestive of cirrhosis. He was compound heterozygous for 2332C→T and 2755C→G in the *ATP7B* gene, but was free from mutations in the *HFE* gene. Trientine hydrochloride at 2500 mg/day reduced his serum ceruloplasmin protein from 8.9 to 5.4 mg/dL within 3 months. A liver specimen taken after 3 years of treatment was still positive for histochemical iron and his serum ferritin was 200 ng/mL and serum ceruloplasmin was 2.0 ng/mL, whereas his serum ferroxidase activity was as low as 70 U/L.

Phlebotomy during trientine treatment were carried out after obtaining written informed consent from each patient where the end-point was a serum ferritin concentration of 10 ng/mL or a hemoglobin concentration of 12 g/dL. As shown in Figure 1, postphlebotomy anemia developed in both patients when their serum ferritin levels were 38 and 17 ng/mL, respectively.

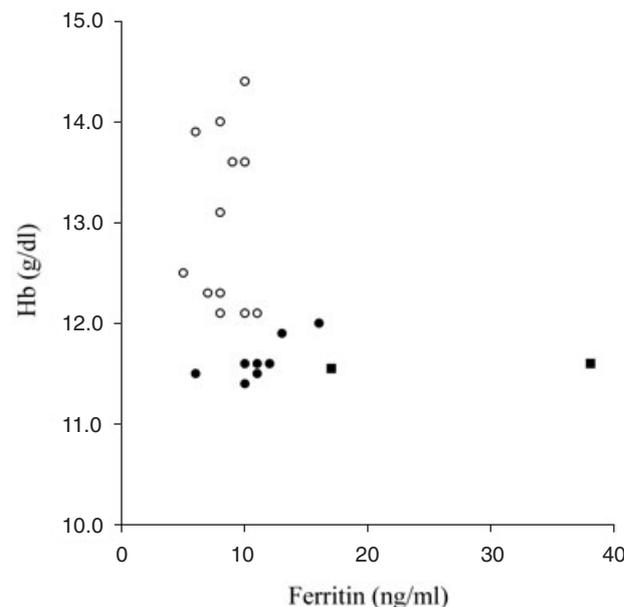


Figure 1 The relationship between hemoglobin and serum ferritin levels as end-points for phlebotomy. Postphlebotomy anemia developed in two patients with Wilson's disease when their serum ferritin levels reached 38 and 17 ng/mL, respectively. Eight patients with chronic hepatitis C (CHC) did not complete iron removal because of the early onset of anemia. The highest serum ferritin level was 16 ng/mL at the end-point caused by anemia for these patients. Twelve patients with CHC tolerated phlebotomy without developing anemia. (■) Wilson's disease; (●) CHC with incomplete iron removal; (○) CHC with complete iron removal.

Compared to patients with chronic hepatitis C, their end-points were high, suggesting their intolerance to iron removal therapy.

Ferroxidase deficiency as a result of hypoceruloplasminemia might be a specific mechanism involved in iron deposition associated with impaired iron transport caused by Wilson's disease. This hypothesis is supported by the observation that iron removal by phlebotomy was insufficient for a patient with aceruloplasminemia.² Chelation therapy using penicillamine or trientine might result not only in the effective excretion of toxic copper, but also in the inhibition of internally bioavailable copper. When iron overload is a side-effect of using these chelators, discontinuation of the drugs might not be recommended, but phlebotomy is enough to control iron-induced oxidative stress.⁵ In future, the effects of copper chelation on iron metabolism should be studied using a larger number of patients.

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REFERENCES

- 1 Attieh ZK, Mukhopadhyay CK, Seshadri V, Tripoulas NA, Fox PL. Ceruloplasmin ferroxidase activity stimulates cellular iron uptake by a trivalent cation-specific transport mechanism. *J. Biol. Chem.* 1999; **274**: 1116–23.
- 2 Hellman NE, Schaefer M, Gehrke S *et al.* Hepatic iron overload in aceruloplasminemia. *Gut* 2000; **47**: 858–60.
- 3 Walshe JM. Treatment of Wilson disease with penicillamine. *Lancet* 1960; **i**: 188–92.
- 4 Walshe JM. Treatment of Wilson disease with trientine (trienthylene tetramine) dihydrochloride. *Lancet* 1982; **i**: 643–7.
- 5 Shiono Y, Wakusawa S, Hayashi H *et al.* Iron accumulation in the liver of male patients with Wilson disease. *Am. J. Gastroenterol.* 2001; **96**: 3147–51.

ESOPHAGEAL METASTASIS OF HURTHLE CELL THYROID CARCINOMA EIGHT YEARS AFTER A SUBTOTAL THYROIDECTOMY THAT MIMICKED ESOPHAGEAL HEMANGIOMA

To the Editor,

Malignant tumors of the upper gastrointestinal tract are relatively rare and can be seen as a result of direct invasion or metastasis. Neck and head tumors, malignant melanoma, lung and breast cancer are the most common metastatic tumors that involve the esophagus. Herein, we describe a patient presenting with hemate-

mesis, dysphagia and odynophagia because of esophageal metastasis of relapsing Hurthle cell thyroid carcinoma 8 years after a subtotal thyroidectomy.

A 69-year-old man was admitted to hospital with hematemesis. He had no weight loss but he had experienced proximal dysphagia and odynophagia for 1 month. He had a left lobe hepatectomy because of hepatic hemangioma 20 years earlier and a subtotal thyroidectomy because of multiple thyroid nodules 8 years earlier. He smoked 90 packets of cigarettes a year. There was no obvious pathological feature except mild pallor on physical examination.

Upper endoscopy showed a 3 × 4 cm, dark red colored, intraluminal polypoid mass which was mimicking hemangioma and stromal tumors. A biopsy could not be carried out because of the suspicion of hemangioma. Neck and thorax magnetic resonance imaging (MRI)

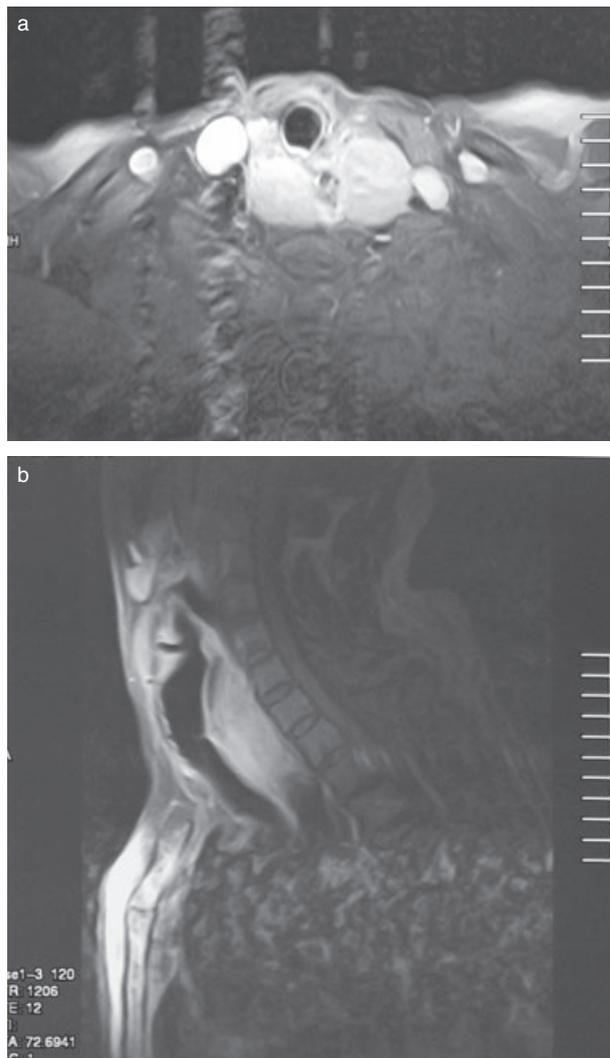


Figure 1 Fat saturated axial and sagittal T1 weighted plane images show mass lesion around the esophageal lumen. (a) This tumoral lesion has a lobulated contour and homogenous contrast enhancement after Gd-DTPA injection. (b) The mass lesion is seen around the esophageal lumen and on the posterior of the trachea with compression effect.

showed an external compression to the esophagus caused by 32×27 mm solid mass in the upper part of the thorax. The mass was extending cranially, it was well arranged with homogenous intensity with two nodules which were 37 and 36 mm in diameter. The nodules were thought to be originating from the thyroid gland and also involving the retrosternal part of the mediastinum. The nodules were causing remarkable compression to the esophageal lumen (Fig. 1a,b). At first, the patient was thought to have esophageal hemangioma, however, because MRI images pointed to the thyroid nodules, thyroid carcinoma could not be ruled out. There was no activity on thyroid scintigraphy. Then the patient was diagnosed with thyroid carcinoma, and total thyroidectomy and segmental resection of the esophageal metastasis were carried out. Histopathological examination showed Hurthle cell carcinoma of both the thyroid nodules and the resection material of the esophagus. The patient was followed up for 5 months in outpatient clinics, he was asymptomatic with normal esophagography.

Locally advanced thyroid carcinoma involving the esophagus originates from direct invasion of the tumor and is usually confined to the muscularis without extension into submucosa or mucosa layers. Patients complain of dysphagia and odynophagia resulting from direct invasion of the esophagus or extrinsic compression in association with laryngotracheal invasion.¹ The patient in the present study had dysphagia, odynophagia and hematemesis. At first, it was thought to be esophageal hemangioma because he had a left liver lobectomy as a result of a giant hemangioma and we did not attribute it to thyroid carcinoma recurrence. However, surgical resection and pathological examination showed Hurthle cell carcinoma.

As with esophageal involvement of locally advanced thyroid carcinoma such as Hurthle cell carcinoma, shave resection to en bloc resection, segmental resection and esophagectomy can be carried out. If surgical treatment can not be applied because of extensive involvement, palliative dilation or stent placement might be helpful for symptomatic patients.¹ In the present

patient, total thyroidectomy and segmental esophageal resection were carried out.

Metastatic tumors of the upper gastrointestinal tract, especially the of the upper part of the esophagus, are relatively rare.^{2,3} If esophageal metastasis is found, neck and head tumors, malign melanoma, lung and breast cancer must be considered.¹⁻³ In addition to this, hepatocellular carcinoma, osteogenic sarcoma, renal cell carcinoma, germ cell tumors and prostate cancer are the other metastatic tumors to the esophagus that were published as case reports.²⁻⁵

In conclusion, metastatic cancer to the esophagus is a rare occurrence. The diagnosis should be considered when patients have dysphagia and odynophagia or alarming symptoms if they have a history of previous primary cancer.

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REFERENCES

- 1 Kebebew E, Clark OH. Locally advanced differentiated thyroid cancer. *Surg. Oncol.* 2003; **12**: 91-9.
- 2 Simchuk EJ, Low DE. Direct esophageal metastasis from a distant primary tumor is a submucosal process: a review of six cases. *Dis. Esophagus* 2001; **14**: 247-50.
- 3 Kadakia SC, Parker A, Canales L. Metastatic tumors to the upper gastrointestinal tract: endoscopic experience. *Am. J. Gastroenterol.* 1992; **87**: 1418-23.
- 4 Cho A, Ryu M, Yoshinaga Y *et al.* Hepatocellular carcinoma with unusual metastasis to the esophagus. *Hepatogastroenterology* 2003; **50**: 1143-5.
- 5 Nakamura T, Mohri H, Shimazaki M *et al.* Esophageal metastasis from prostate cancer: diagnostic use of reverse transcriptase-polymerase chain reaction for prostate-specific antigen. *J. Gastroenterol.* 1997; **32**: 236-40.