

# Evaluating the trace elements in preoperative and postoperative duration of hydatid surgery

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## ABSTRACT

**الأهداف:** الهدف من الدراسة هو تقييم مستويات مصّل الزنك والمغنيسيوم والنحاس قبل وبعد العملية الجراحية لدى المرضى المصابين بكيس داء المكوّرات المشوكة.

**الطريقة:** أجريت هذه الدراسة على المرضى المصابين بكيس داء المكوّرات المشوكة في الفترة ما بين عام ٢٠٠٠م و٢٠٠٥م بكلية الطب بجامعة كوكوروا بمدينة أدنا بتركيا. تم قياس مستويات مصّل الزنك والمغنيسيوم والنحاس لخمسة وثمانين مريضاً يعانون من كيس داء المكوّرات المشوكة قبل العملية الجراحية وبعدها بعام واحد.

**النتائج:** شملت هذه الدراسة المرضى الذين يعانون من وجود كيس داء المكوّرات المشوكة في الكبد (٨٥ مريضاً، ٤٨ ذكراً) وبالغين سليمين (٤٠ بالغ، ١٧ ذكراً) كمجموعة التحكم. عند المقارنة مع مجموعة التحكم، كانت متوسط مستويات مصّل الزنك والمغنيسيوم أقل وكان متوسط مستويات مصّل النحاس أعلى في الفترة قبل العملية الجراحية. تبين أنه بينما تزيد فترة الأعراض فإن مستويات مصّل الزنك والمغنيسيوم تنخفض وتزداد مستويات مصّل النحاس. تمت مقارنة مستويات مصّل هذه العناصر في الفترة قبل العملية الجراحية ومجموعة التحكم بعد عام واحد بالتساوي مع مستويات مجموعة المرضى.

**خاتمة:** إن احتمالية زيادة استهلاك الزنك والمغنيسيوم وإفراز النحاس من قبل الطفيليات قد يؤدي الى تقييم متابعة الجراحية العدارية بالإضافة الى الطرق الإشعاعية والمصلية.

**Objective:** To evaluate the preoperative and postoperative values of serum levels of zinc, magnesium, and copper in patients with cystic echinococcosis (CE).

**Methods:** This study was conducted on patients with CE between 2000-2005 at the Faculty of Medicine, University of Cukurova, Adana, Turkey. Serum levels of zinc, copper, and magnesium of 85 patients with CE were measured both before and one year after the operation. Patients with liver CE (85 patients, 48 males) and healthy adults (40 adults, 17 males) as the control group were enrolled in the study.

**Results:** When compared with the control group, the mean serum levels of zinc and magnesium were lower and the mean serum level of copper was higher in the preoperative period. It was found that as duration of symptoms increased, serum zinc and magnesium levels decreased and copper levels increased. The serum levels of these elements in postoperative and control patients after one year compared equally with the levels in the initial control.

**Conclusion:** The probability of increased consumption of zinc and magnesium and secretion of copper by the parasite may lead to evaluating the follow up of hydatid surgery in addition to radiological and serological methods.

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Cystic echinococcosis (CE) is accepted to be the most important parasitic zoonosis in the Mediterranean countries causing serious problems to public health and national economy.<sup>1,2</sup> It can maintain its fertility against the immune status of the body and occur without any symptoms for long periods.<sup>3,4</sup> The relationship of trace elements and certain disease groups such as cancer, immune deficiencies, tuberculosis, and rheumatoid diseases have been well identified.<sup>5-7</sup> However, there are only 3 published studies in recent years,<sup>2</sup> demonstrating the relationship of trace elements with CE,<sup>8,9</sup> and the other with Alveolar Echinococcosis (AE).<sup>10</sup> The exact mechanism responsible for alterations in trace

element levels in patients with chronic inflammatory or infectious diseases is largely unclear and requires further evaluation.<sup>5-7,10</sup> In many studies, it was reported that the cause could be the acute phase reaction due to the host response against inflammation.<sup>5-8</sup> The aim of this study is to demonstrate the relationship between CE and serum levels of zinc (Zn), magnesium (Mg), and copper (Cu) with respect to parasite fertilization, cyst dimension, and duration of the diseased period, and consequently to assess alterations after treatment.

**Methods.** Patients with liver CE (85 patients, 48 males) treated between 2000 and 2005 at the Department of General Surgery, Faculty of Medicine, University of Cukurova, Adana, Turkey and healthy adults (40 adults, 17 males) as the control group were enrolled in the study. The study protocol was reviewed and approved by the Faculty of Medicine Ethics Committee and an informed consent were obtained. Preoperative diagnosis was established by radiologic (US and CT) and serologic (Western blotting) methods. Anti-*Echinococcus granulosus* (*E. granulosus*) antibodies were determined by Echinococcosis Western blotting, Euroimmun, Germany, according to the protocols established by the manufacturer. The monitoring period for all patients was 12 months. Demographic data, symptom duration, cyst localizations, count and diameters of cysts were recorded. For multiple cysts, cyst dimension was described as the diameters of all cysts. All patients were classified according to the WHO- Informal Working Group on Echinococcosis (WHO-IWGE) classification of cystic hydatid disease.<sup>11</sup> Stages 1, 2, and 3 were accepted as the more fertile group, stages 4 and 5 were accepted to be the degenerated and calcified group. Total cystectomy was performed in suitable cases; if not suitable, partial cystectomy and capitonnage methods were used. Cyst cavities were controlled with a laparoscopic camera for eliminating the bile fistula. While right lobe cavities were filled with omentum (omentoplasty), left lobe cavities were filled with falciform ligament flap. Serum Zn, Cu, and Mg levels were measured preoperatively and after 12 months following the operation. To minimize the effect of hydration alterations of daytime on results, serum specimens were taken from all participants at exactly the same time of the day (8:00 am). All CE patients were administered albendazole 10 mg/kg/day as post-operative medical treatment for 6 months, in the sequence of "3 weeks with and one week without" medication. No side effects were observed in the study group during the 6 months. Patients were examined by a combination of radiologic and serologic methods at the end of the 12th month. No patient was treated according to postoperative serum levels of

trace elements and none were given a specific diet. All patients operated with CE were included in the study, however, the patients that could not be monitored and refused postoperative chemotherapy were excluded from the study. A total of 95 cases of hydatid disease were operated in our department in the study period. Five patients with no informed consent were not included in the study. In the postoperative first year, western-blotting was positive and relapse suspicion was detected by radiologic investigations in 3 cases. However, it was found out by inquiry that these 3 cases, due to sociocultural circumstances, had not taken the albendazole medication. Considering possible drawbacks such as standardization impairment, these 3 cases were also excluded from the study. Another patient who was serologically and radiologically doubtful for relapse was recommended confirmation of diagnosis by detection of serum trace element levels and treatment with puncture, aspiration, injection, re-aspiration [PAIR] method. However, this patient also, when refusing both recommendations, was excluded from the study. The PAIR was performed and relapse was confirmed only in one suspicious case. In order to measure Cu, Zn, and Mg levels, serum was separated from the clotted blood and stored in acid-washed tubes at -20°C until the analysis by atomic absorption spectrometry (AAS). Specimens were diluted with de-ionized water at ratios of 1:1, 1:5 and 1:50, for the determination of Cu, Zn, and Mg. Calibration standards and quality control specimens were prepared in the same way. Readings for Cu, Zn, and Mg were performed at wave lengths of 324.8, 213.9, and 285.2 nm.<sup>12,13</sup>

Statistical analysis was performed using the statistical package SPSS Version 12.0. For each continuous variable, normality was checked by Shapiro Wilks test. Comparisons of continuous variables were applied using the student t test and Pearson correlation test. The categorical variables between the groups were analyzed using the Chi square test. Data were expressed as mean  $\pm$  SD (min-max), number (percentage) and a p-value <0.05 which is considered significant.

**Results.** The demographic data are presented in Table 1. The gender and age characteristics of the control group were similar to the CE group. Cysts were mostly located on the right lobe (52.9%). The postoperative first year recurrence rates were 5.2% for suspicious recurrence and 1.05% for definite diagnosis. The mean serum values of Cu, Zn, and Mg in the CE group and the control group are presented in Table 2. While the mean serum levels of Zn and Mg were lower, the mean serum level of Cu was higher in the CE group when compared with the control group (p<0.001). The mean preoperative and postoperative serum values of Cu, Zn,

and Mg in the CE group are also presented in Table 2. The mean preoperative serum levels of Zn and Mg were lower, but the mean serum Cu level was higher when compared with postoperative measurements ( $p < 0.001$ ). No statistical difference was observed when the postoperative serum levels of Cu, Zn, and Mg were compared with the control group. The mean serum levels of Cu, Zn, and Mg in CE groups according to the WHO classification are presented in Table 3. We found that the mean serum Cu level was significantly higher in stages 1, 2, and 3 of CE, which are considered to be the most fertile stages, than in stages 4, and 5 of CE, in which the cysts are considered as de-generated and calcified. The mean serum Zn level was significantly lower than in the de-generated and calcified group when compared with the most fertile group ( $p < 0.001$ ). However, the mean serum Mg level was not significantly different between

both groups. The mean value of Zn, Cu, and Mg were significantly different from the control groups both in the most fertile and de-generated and calcified groups (Figure 1). When cyst dimension and symptom duration were compared with serum Zn, Cu, and Mg levels, it was found that as the symptom duration increased, serum Zn ( $r = -0.32$ ) and Mg ( $r = -0.33$ ) levels decreased, but mean serum Cu ( $r = 0.23$ ) levels increased ( $p < 0.05$ ).

**Discussion.** In this study, we found that the mean serum levels of Zn and Mg decreased, the mean serum Cu level increased in patients with CE in the preoperative period when compared with healthy adults. In addition, the serum levels of these trace elements in the postoperative period were similar when compared with healthy adults. The relationship between trace elements and many disease groups such

**Table 1** - Subject characteristics.

Characteristics	Control group (n=40)	CE group (n=85)	P-value
Age* (years)	50.1 ± 10.9* (28-68)†	46.9 ± 8.7* (28-64)†	0.08
<i>Gender</i>			0.9
Male	17	48	
Female	23	37	
Cyst dimension cm*	-	11.5 ± 6.6* 4-27†	
Count of cysts*	-	20 ± 1.3* 1-5†	
Symptom duration (month)*	-	23.2 ± 16.3* 3-60†	
<i>Cyst localization (n [%])</i>			
Right hepatic lobe		45 (52.9)	
Left hepatic lobe	-	11 (12.9)	
Both		29 (34.1)	

\*mean ± SD, †minimum-maximum

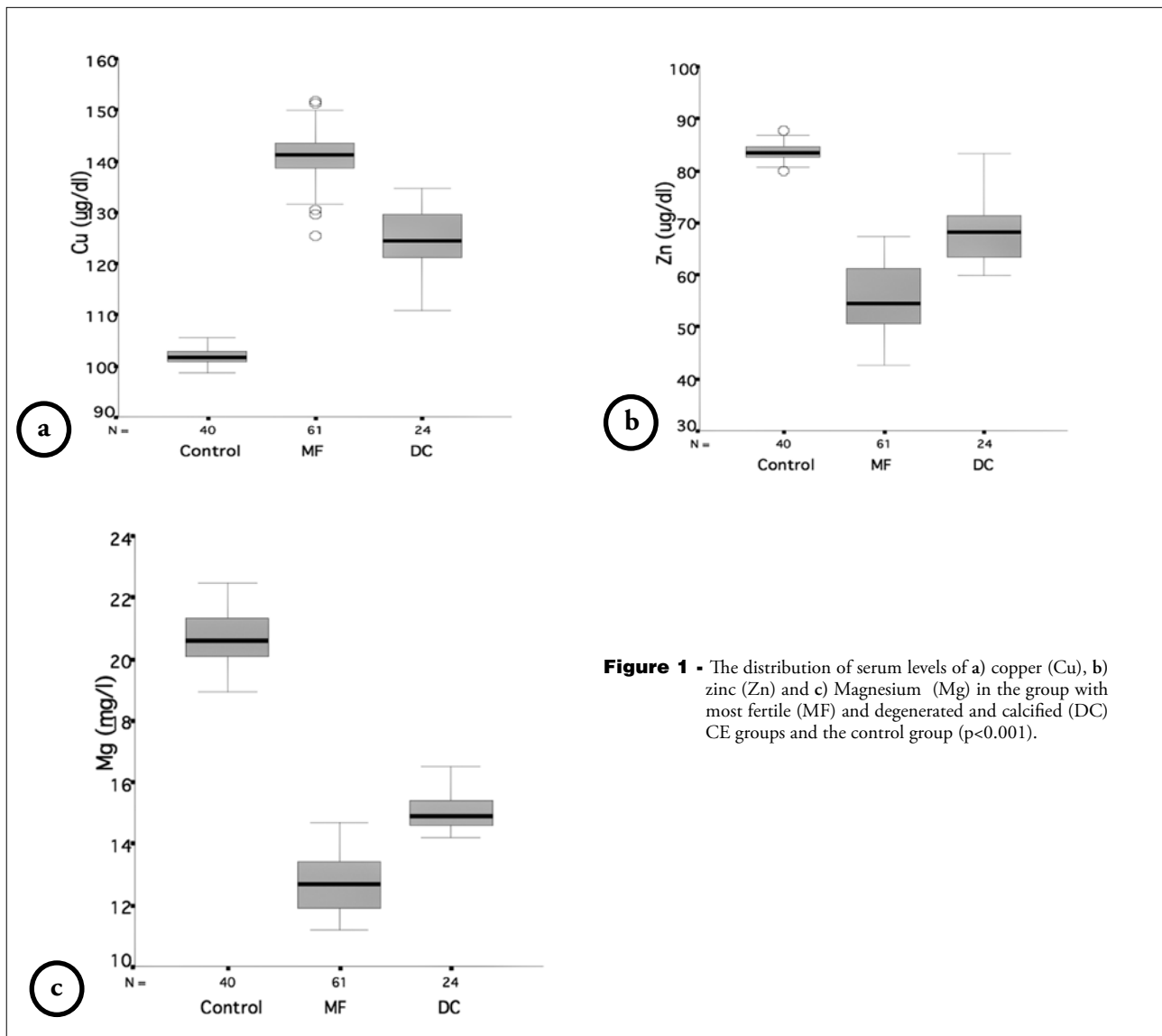
**Table 2** - The distribution of serum levels of copper (Cu), zinc (Zn) and magnesium (Mg) in the group with pre-operative and post-operative cystic echinococcosis (CE) and the control group.

Elements	Control group n=40	CE group		P-value		
		Pre-operative (n=85)	Post-operative (n=85)	Between control and preoperative	Between preoperative and postoperative	Between control and postoperative
Cu (µg/dl)	101.9 ± 1.5	136.1 ± 9.09	101.7 ± 1.4	0.0001	0.0001	0.539
Zn (µg/dl)	83.5 ± 1.6	59.2 ± 9.29	83.4 ± 1.9	0.0001	0.0001	0.517
Mg (mg/l)	20.7 ± 0.8	13.3 ± 1.32	20.8 ± 1.2	0.0001	0.0001	0.512

**Table 3** - The mean serum values of copper (Cu), zinc (Zn) and magnesium (Mg) in cystic echinococcosis (CE) group according to the World Health Organization (WHO) classification.

Groups/WHO classification	Zn (µg/dl) Mean±SD	C (µg/dl) Mean±SD	Mg (mg/l) Mean±SD
<i>Most fertile (n=61)</i>	57.36 ± 6.48	139.8 ± 2.66	12.91 ± 0.61
CE1 (n=10)	63.51 ± 2.97	137.55 ± 5.66	13.56 ± 0.92
CE2 (n=30)	50.85 ± 3.61	142.74 ± 5.69	12.33 ± 0.85
CE3 (n=21)	57.99 ± 5.96	139.11 ± 3.8	12.85 ± 0.61
<i>De-generated and calcified (n=24)</i>	62.26 ± 3.81	126.7 ± 5.09	14.85 ± 0.86
CE4 (n=17)	78.24 ± 7.92	120.05 ± 5.35	15.07 ± 0.67
CE5 (n=7)	71.57 ± 6.5*	123.46 ± 3.3*	14.95 ± 0.11*

\**p*<0.0001 between most fertile and de-generated and calcified group



**Figure 1** - The distribution of serum levels of a) copper (Cu), b) zinc (Zn) and c) Magnesium (Mg) in the group with most fertile (MF) and degenerated and calcified (DC) CE groups and the control group (*p*<0.001).

as cancer, immune deficiencies, tuberculosis, chronic hemodialysis patients, and rheumatoid diseases have been identified in the literature.<sup>5-7</sup> However, there are only 2 published studies in recent years, which demonstrate the relationship between trace elements and CE,<sup>8,9</sup> and one study with AE.<sup>10</sup> Ozen et al,<sup>8</sup> in a study which included 12 patients and 12 sheep with CE found a decrease of selenium and Zn levels and an increase of Cu levels. Chowdhury and Singh<sup>9</sup> reported the relationship between trace elements and diameter and wall thickness of cysts in buffalo. Wellinghausen et al<sup>10</sup> found that Zn level was lower in the group that includes 40 patients with AE when compared with the control group. The symptom duration and fertility were correlated with the levels of trace elements in this study. Because of the difficulty in defining the life span of the disease, the symptom duration was accepted to be the life span. It was found that with symptom duration, Zn and Mg levels decreased and Cu level increased. However, no study has been published that identifies the correlation between trace elements and the life span of disease. Chowdhury and Singh<sup>9</sup> reported the relationship between trace elements in the cyst fluid and diameter of cysts in buffalo. However, no significant relationship was found between serum trace elements and cyst dimension in this study. When the level of trace elements was analyzed in accordance with the classification defined by the WHO study group, a correlation was found between the level of trace elements and the natural course of the hydatid cyst.<sup>11</sup> According to the present study, serum Cu levels were significantly higher at stages 1, 2, 3 of CE (the most fertile stages) and the Zn levels were significantly lower at the stages 4, 5 (the de-generated and calcified stages) of the disease. However, the serum Mg level was not significantly different between both stage groups. Ozen et al<sup>8</sup> found that cyst fluid in sheep has a more significant relationship with serum trace elements than it has in humans. This might be caused by cysts being more fertile in sheep, which suggests a possible relationship of fertile capacity with trace element levels. Wellinghausen et al<sup>10</sup> reported a negative correlation between progressive disease and serum zinc levels in patients with AE. The exact mechanism responsible for alterations in trace element levels in patients with chronic inflammatory or infectious diseases is largely unclear and requires further evaluation.<sup>5-7,10</sup> In many studies, it was reported that the cause could be the acute phase reaction due to host response against inflammation.<sup>5-8</sup> Wellinghausen et al<sup>10</sup> suggested that the explanation of the decrease in the Zn level of patients with AE could be due to the increased consumption of zinc by the growing parasite. As there is a correlation with symptom duration and fertility in our study, we think that these findings probably resulted from

consumption of Zn and Mg and secretion of Cu into the circulation by the parasite. The follow-up of HD surgery is an important issue, due to relapse or residue problems after hydatid disease surgery.<sup>14-16</sup> Especially in the first postoperative year, cyst like structures are usually seen in the surgical evacuation site, but differential diagnosis of these structures (whether as persistent changes or as newly formed parasites) is always difficult.<sup>15-18</sup> Results of serologic tests remain positive long after operation, and only demonstration of the scolices in the remaining cavity ensures definitive diagnosis.<sup>18,19</sup> Therefore, radiological methods (ultrasonography and computer tomography) and serologic tests are insufficient for follow-up of hydatid disease in the first postoperative year.<sup>14,19,20</sup> In our study, it was established that the serum trace element levels in treated patients were normal in the postoperative first year. With these results, we suggest that trace element levels, in addition to radiologic and serologic methods, can be useful for follow up in the postoperative course.

In conclusion, the decrease of Zn and Mg levels and the increase of Cu levels were established in the serum of patients with CE. A statistically significant relation was found between these alterations and symptom duration. While direct correlation between Zn levels and negative correlation between Cu levels and fertility were detected, no association was established between Mg levels and fertility. These findings imply the probability of increased consumption of Zn and Mg and secretion of copper by the parasite in addition to acute phase reaction for etiopathogenesis. Trace elements return to their normal level in the 12th month after operation. Therefore, the analysis of trace element levels could be valuable for the follow up of hydatid surgery in addition to radiological and serological methods.

## References

1. Eckert J, Deplazes P. Biological, epidemiological, and clinical aspects of echinococcosis, a zoonosis of increasing concern. *Clin Microbiol Rev* 2004; 17: 107-135.
2. Seimenis A. Overview of the epidemiological situation on echinococcosis in the Mediterranean region. *Acta Trop* 2003; 85: 191-195.
3. Koltas IS, Yucebilgic G, Bilgin R, Parsak CK, Sakman G. Serum malondialdehyde level in patients with cystic echinococcosis. *Saudi Med J* 2006; 27: 1703-1705.
4. Yoshikawa M, Hirai T, Ouji Y, Marugami NA, Toyohara M, Nishiofuku M, et al. Hydatid cysts in the liver. *Intern Med* 2006; 45: 565-566. Epub 2006 15.
5. Rivera MT, De Souza AP, Araujo-Jorge TC, De Castro SL, Vanderpas J. Trace elements, innate immune response and parasites. *Clin Chem Lab Med* 2003; 41: 1020-1025.
6. Jain A, Mukherjee A, Chattopadhyaya D, Saha K. Biometals in skin and sera of leprosy patients and their correlation to trace element contents of *M. leprae* and histological types of the disease; a comparative study with cutaneous tuberculosis. *Int J Lepr Other Mycobact Dis* 1995; 63: 249-258.

7. Kassu A, Yabutani T, Mahmud ZH, Mohammad A, Nguyen N, Huong BT, et al. Alterations in serum levels of trace elements in tuberculosis and HIV infections. *Eur J Clin Nutr* 2006; 60: 580-586.
8. Ozen N, Celik C, Ozkan K, Malazgirt Z, Isimer A, Sayal A. Trace elements in hydatid disease. *J Trace Elem Electrolytes Health Dis* 1992; 6: 67-70.
9. Chowdhury N, Singh R. Distribution of some elements in hydatid cysts of *Echinococcus granulosus* from buffalo (*Bubalus bubalis*). *J Helminthol* 1993; 67: 112-114.
10. Wellinghausen N, Jochle W, Reuter S, Flegel WA, Grunert A, Kern P. Zinc status in patients with alveolar echinococcosis is related to disease progression. *Parasite Immunol* 1999; 21: 237-241.
11. Sayek I, Tirnaksiz MB, Dogan R. Cystic hydatid disease: current trends in diagnosis and management. *Surg Today* 2004; 34: 987-996.
12. Butrimovitz GP, Purdy WC. The determination of zinc in blood plasma by atomic absorption spectrometry. *Anal Chim Acta* 1977; 94: 63-73.
13. Fernandez FJ, Kahn HL. Clinical methods for atomic absorption spectroscopy. *Clin Chem Newsl* 1971; 3: 24-26.
14. Kapan M, Kapan S, Goksoy E, Perek S, Kol E. Postoperative recurrence in hepatic hydatid disease. *J Gastrointest Surg* 2006; 10: 734-739.
15. Sielaff TD, Taylor B, Langer B. Recurrence of hydatid disease. *World J Surg* 2001; 25: 83-86.
16. Silva MA, Mirza DF, Bramhall SR, Mayer AD, McMaster P, Buckels JA. Treatment of hydatid disease of the liver. Evaluation of a UK experience. *Dig Surg* 2004; 21: 227-233.
17. Paksoy M, Karahasanoglu T, Carkman S, Giray S, Senturk H, Ozcelik F, et al. Rupture of the hydatid disease of the liver into the biliary tracts. *Dig Surg* 1998; 15: 25-29.
18. Lawn SD, Bligh J, Craig PS, Chiodini PL. Human cystic echinococcosis: evaluation of post-treatment serologic follow-up by IgG subclass antibody detection. *Am J Trop Med Hyg* 2004; 70: 329-335.
19. Force L, Torres JM, Carrillo A, Busca J. Evaluation of eight serological tests in the diagnosis of human echinococcosis and follow-up. *Clin Infect Dis* 1992; 15: 473-480.
20. Bozkurt B, Soran A, Karabeyoglu M, Unal B, Coskun F, Cengiz O. Follow-up problems and changes in obliteration of the residual cystic cavity after treatment for hepatic hydatidosis. *J Hepatobiliary Pancreat Surg* 2003; 10: 441-445.

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Abdo AA, Azzam NA, Al-Faleh FZ, Al-Jebreen AM, Al-Mofleh IA, Al-Amri SM, Al-Rashid RS, Al-Freih HM, Habbal TA, Al-Swat KE, Isnani AC, Alsheikh A, Ahmed SS. Histological and laboratory features of patients undergoing liver biopsy at a university hospital in Central Saudi Arabia. *Saudi Med J* 2006; 27: 1493-1497.

Helvaci MR, Algin MC, Ozdogu H. Follow-up of hepatitis C virus infected patients. *Saudi Med J* 2006; 27: 1429-1431.