



## Original Article

# Coinfection of *Toxoplasma Gondii* and Intestinal Parasites among Elderly Immunocompromised Patients

Mohammad Reza Mahmoudi <sup>\*1,2,3</sup> , Behnaz Rahmati <sup>3</sup>

<sup>1</sup> Cellular and Molecular Research Center, Faculty of Medicine, Guilan University of Medical Sciences, Rasht, Iran

<sup>2</sup> Medical Biotechnology Research Center, School of Paramedicine, Guilan University of Medical Sciences, Rasht, Iran

<sup>3</sup> Department of Parasitology, Faculty of Medicine, Guilan University of Medical Sciences, Rasht, Iran

\* **Corresponding Author:** Department of Parasitology, Faculty of Medicine, Guilan University of Medical Sciences, Rasht, Iran. **Tel:** +989126220613, **Email address:** [mrmahmoodi2002@yahoo.com](mailto:mrmahmoodi2002@yahoo.com)

## ABSTRACT

### Article history

Received 16 Jun 2019

Accepted 12 Apr 2020

**Citation:** Mahmoudi MR, Rahmati B. Coinfection of *Toxoplasma gondii* and intestinal parasites among elderly immunocompromised patients. *Elderly Health Journal*. 2020; 6(1): 31-35.

**Introduction:** Patients with cancer and hemodialysis are prone to opportunistic infections. The present research aims to examine the frequency of Toxoplasmosis and intestinal parasites in these patients.

**Methods:** In the present cross-sectional study, seventy stool and blood samples from immunocompromised patients (46 hemodialysis and 24 cancer patients) from Guilan Province were collected in 2017-2018. Different tests including direct smear examination and formalin-ether was conducted for the intestinal protozoan parasites. To detect coccidian parasites (e.g. *Cryptosporidium* spp.) Ziehl Neelsen staining was applied. The anti *T.gondii* antibodies were detected via ELISA method.

**Results:** Totally 6.52% and 4.16% cases of hemodialysis and cancer patients were positive for intestinal parasite infection respectively. *Blastocystis hominis* was only intestinal parasite that were detected in these patients. IgG anti-Toxoplasma antibody detected in 35 (76.8%) hemodialysis and 15 (62.5%) cancer patients. IgM antibody were found in 1 elderly hemodialysis patient. Mix infection (Toxoplasmosis and *Blastocystis*) was observed in 6.52% and 4.16% in hemodialysis and cancer patients respectively.

**Conclusion:** It is recommended to evaluate these patients for intestinal parasitic infections during hemodialysis or chemotherapy sessions. Data showed the high percentage of elderly patients were susceptible to reactivation of chronic Toxoplasmosis. Therefore, in order to refer them for early therapy or other interventions, it is important that elderly hemodialysis and cancer patients with toxoplasma infection be diagnosed and identified.

**Keywords:** Aged, Immunocompromised Host, Parasites, Toxoplasmosis

## Introduction

Age-related physical disabilities, illness and reduced self-care ability at an older age have led to a poor personal hygiene so the sensitivity to infections will increase in the elderly (1). Cancer and End-stage Renal Disease are among the diseases that become more likely as we age (2, 3). One of consequences of aging is a decrease in immune function (4). Parasitic infections are a health problem in all societies (5) and older persons

generally have a greater susceptibility to infections than younger adults. It is well-known that aging is associated with immune dysfunction, especially in cell-mediated immunity (6). Patients with some type of immunocompromised condition and those submitted to immunosuppressive therapy are more likely to be infected with parasites (7).

Intestinal parasites are worldwide distribution, and elderlies are more susceptible to these diseases than

young individuals (8). Intestinal parasitic infections that cause auto-limited diarrhea in immunocompetent patients may cause severe diarrhea in immunocompromised individuals (7).

Toxoplasmosis is a prevalent zoonotic infection in warm-blooded creatures. Once oocysts and tissue are ingested, infection arises (9). Toxoplasma infection is mainly asymptomatic; however, in immunosuppressed patients, e.g. individuals with AIDS, cancer patient or organ transplant recipients, can cause severe diseases such as encephalitis or systemic infections (10-12). The disease can be due to acquired infection or latent infection reactivation (13). Most importantly, the chronic Toxoplasmosis reactivation in seropositive hemodialysis patients may cause acute disease with fatal outcomes (14, 15). Thus, the Toxoplasmosis diagnosis is central in elderly hemodialysis and cancer patients (16, 17). Despite the word population aging, a few studies are on the parasitic infection prevalence. The study aimed to examine the frequency of intestinal protozoa and Toxoplasma in elderly immunocompromised residents in Guilan-north of Iran.

## Methods

### *Study design and sampling*

This cross-sectional research was conducted between 2017 and 2018 in Guilan province. This province has a humid subtropical climate with the heaviest rainfall, around 1,400 mm. The humidity can increase to 90 percent in summer, and humid environments are suitable condition for Toxoplasma survival (18).

The samples of stool and blood were obtained from 70 elderly immunocompromised patients (46 hemodialysis and 24 cancer patients) of both genders (41 males and 29 female), with the age between 60 - 78 years old. The epidemiological data, including age, gender, and clinical signs and symptoms of some cases were also obtained through a predesigned questionnaire by an expert laboratory technician for each individual.

### *Detection of intestinal parasites*

All stool specimen were tested via direct wet mount (both the saline and iodine wet mount) technique and formalin-ether sedimentation technique under microscopic observation (Zeiss, Germany, 40 X magnifications). In this research to detect coccidian parasites such as *Cryptosporidium* spp., Ziehl Neelsen acid-fast stain staining were employed (19). Per person, three slides were created and studied by trained professional staff.

### *Serological evaluation for Toxoplasmosis*

For the serum separation, five ml of venous blood specimen were gained and then kept at  $-20^{\circ}\text{C}$ . A commercial ELISA kit was used to detect IgG and IgM antibodies to *T. gondii* (Toxo IgG and Toxo IgM Euroimmun). Based on the recommendations of the producer, IgG absorbance levels  $< 9$  were regarded to be negative, 9-11 borderline, and  $> 11$  positive; IgM absorbance levels  $< 0.9$  were assumed to be negative, 0.9-1.1 borderline, and  $> 1.1$  positive.

### *Statistical analysis*

SPSS version 16 software was applied to analyze the data.

### *Ethical considerations*

The Ethical Research Committee, Guilan University of Medical Sciences approved this research with ethics number of IR.GUMS.REC.1396.164

## Results

In total, we enrolled 46 hemodialysis patients (group 1) and 24 cancer patients (group 2) including 41 males and 29 females. In hemodialysis patients, the ages ranged from 60 to 78 years old and in the cancer patients from 60 to 77 years old. Men were principal in patients' groups, (58.3% in group 1 and 58.6% in group 2). The intestinal protozoa were found in 6.5% and 4% in elderly hemodialysis and cancer individuals respectively. *Blastocystis hominis* was an only intestinal parasite that was found in these patients. None of fecal samples were diarrheic and all the infected subjects were asymptomatic. Toxoplasma antibody detection test showed that 50 patients were seropositive for Toxoplasma gondii IgG antibodies, including 35 (76.8%) and 15 (62.5%) hemodialysis and cancer patients respectively. IgM antibody was found in one elderly hemodialysis patient. Mix infection (Toxoplasmosis and Blastocystis) was observed in 6.52% and 4.16% in hemodialysis and cancer patients, respectively.

## Discussion

Epidemiological investigations are required to distinguish the demographic of the elderly, as well as their demands for health and social services, which guarantee their life quality improvement (20). Chemotherapy and hemodialysis patients are two groups of immunocompromised subjects exposed to severe effects of parasitic diseases.

Generally, Toxoplasmosis seroprevalence differ from 0% to 95% (21, 22) and is relatively high in the north of Iran (74.6%) (23). In our previous research, Toxoplasma gondii was detected in the certain river water of Iran (9). For Toxoplasma IgG antibodies, approximately 76.8% of hemodialysis and 62.5% of cancer patients were confirmed to be positive in the current work.

The result of the present research indicated a high Toxoplasmosis prevalence in hemodialysis patients, that is similar to findings in Turkey (76.5%) (24), but, higher than other investigations reported in Iran (60.0%), Malaysia (51%), Mexico (10.0%) and Egypt (60%) (25-28).

The latent Toxoplasmosis prevalence was investigated in malignant patients were (45.2%) in Iran (27), 60 (60.0%) in Turkey (29). It was 33% in the non demented elderly group, 41% in the old Alzheimer disease group (30) and 12 % of the elderly in Mexico City (31). Thus, the current work discovered that the Toxoplasmosis prevalence was more than the mean of prevalence in immunodeficiency group in Iran and world. In these

special groups and ages, the prevalence of IgG+ is a potential risk for the reactivation of latent Toxoplasmosis because whenever the body's defensive system gets disturbed, it enables parasite reactivation and becomes an acute infection. On the other side, IgM antibody were found in one elderly hemodialysis patient, so it seems reasonable to take adequate care of such old patients to prevent an acute or latent Toxoplasmosis reactivation in the research area. In the normal people in the northern part of Iran (near the Caspian Sea), the chronic Toxoplasmosis occurrence has been higher compared to other regions in the country (32). This situation may depend upon the appropriate conditions for sporulation, oocysts survival, and transmission to a new host at mild and wet climates. It is known that *T. gondii* infection seroprevalence is high in humid regions (18). Guilan province has a humid climate with a suitable condition for *Toxoplasma* survival in the environment.

The elderly is prone to intestinal parasitic infections (8). In this study, 6.5% and 4.16% of old patients under hemodialysis or chemotherapy were noticed to be infected with intestinal parasites. These findings may correlate with the parasites prevalence among Guilan overall population (33). *Blactocystis* was the one intestinal parasite amongst the infected people in this research (6.52% hemodialysis and 4.16% cancer patients), all being men and asymptomatic. Finding the *Blastocystis* spp as the most intestinal parasites was also in line with other investigations in other age groups (34-36). The frequency of intestinal parasites in old people noticed to be 7.5% (18), 30.5% (8) and 8.72% in Brazil (37). It was, in the current study, lower than that detected in elderly in India (49% of the old people) (38), Brazil (60 + people) (39) and Iran (69 + people) (40). In another study in Brazil, the prevalence of *G. duodenalis*, *Cryptosporidium* spp., *E. histolytica/dispar* in old people was 4.0%, 1.0%, and 0.3% respectively (41). Since the intestinal protozoa *G. duodenalis*, *Cryptosporidium* spp. and *E. histolytica/dispar* are water and food borne disease and related to insufficient hygiene habits, they are significant for old people (42). In earlier studies, these parasites were detected in water samples in Guilan (43-45); however in the current work, all stool samples were negative for the parasites.

The prevalence of intestinal protozoa infections is associated with improper hygiene (46) and climatic conditions (47).

The negativity for some intestinal protozoan parasite demonstrates that the lifestyle and reliable hygiene habits (such as purified drinking water) of the elderly in the research area that causes the decrees of these parasites. Our results show the prevalence of this intestinal parasitic infection in hemodialysis patient was more than the patients under chemotherapy. The cancer patients often have a shorter lifespan and require a higher level of medical care than hemodialysis patients while the latter sometimes spend a 20-year period on hemodialysis and this may cause deficient attention to health care tips and enhance the probability of parasitic infections. Apart from buildings and their equipment, the staff has a key part in transmission or prevention of parasitic infections (48). Hemodialysis patients are frequently exposed to the

infections from nursing personnel, equipment, surfaces and the hands of practitioners (49).

## Conclusions

This study provides new information on the frequency of intestinal parasites and *Toxoplasma* in tow important elderly immunodeficiency groups in Guilan-Iran. The prevalence of Toxoplasmosis was higher in two groups of immunocompromised elderly patients than the many other studies. These immunocompromised patients were potentially susceptible to reactivation of latent Toxoplasmosis which decreases the life quality of these patients. Thus, it would be desirable that the antibody status of patients be known before and during chemotherapy or hemodialysis to refer them to an early therapy or other interventions. In the current research, the only detected protozoan was *B. hominis* that may act as a pathogen in immunocompromised subjects and lead to various types of infections. Thus, periodic parasitology examinations are suggested for hemodialysis and chemotherapy patients. In parallel, inform and educate the patients on how to prevent intestinal parasite and *Toxoplasma* infection.

## Study limitations

Limitations of this research, were sample size and number of stool examination, in which we performed single stool examination, which could have underestimated the parasites prevalence. However, the detection of the invasive stages of protozoa may be missed because of the delays in submitting of some stool samples. This study was unable to assess the possible risk factors related to parasitic infection owing to inadequate demographic, economic and social data.

## Conflicts of interest

No potential conflicts of interest were disclosed.

## Acknowledgments

Authors grateful to hemodialysis and cancer patients, healthy controls, medical doctors and personnel of medical lab for their invaluable cooperation to this project. This project was founded by the Guilan University of Medical Sciences.

## Authors' contributions

MR.M: Technical supports, analysis and interpretation of data, drafting of the manuscript and critical revision of the manuscript for important intellectual content: B.R: contributions to the acquisition of the data. All the authors have read the final manuscript and approved that.

## References

1. El Chakhtoura NG, Bonomo RA, Jump RL. Influence of aging and environment on presentation of

- infection in older adults. *Infectious Disease Clinics*. 2017; 31(4): 593-608.
2. United Nations. *World population Ageing 2007*. New York: United Nations; 2007.
  3. Ghafari M, Sharifirad GR, Zanjani S, Hassanzadeh A. Stress, anxiety and depression levels among elderly referrals to Tehran elderly club. *Salmand: Iranian Journal of Ageing*. 2012; 7(2): 53-9.
  4. Thompson WW, Shay DK, Weintraub E, Brammer L, Cox N, Anderson LJ, et al. Mortality associated with influenza and respiratory syncytial virus in the United States. *The Journal of the American Medical Association*. 2003; 289(2): 179-86.
  5. Alum A, Rubino JR, Ijaz MK. The global war against intestinal parasites—should we use a holistic approach?. *International Journal of Infectious Diseases*. 2010; 14(9):e732-8.
  6. Ben-Yehuda A, Weksler ME. Host resistance and the immune system. *Clinics in Geriatric Medicine*. 1992; 8(4): 701-12.
  7. Botero JH, Castaño A, Montoya MN, Ocampo NE, Hurtado MI, Lopera MM. A preliminary study of the prevalence of intestinal parasites in immunocompromised patients with and without gastrointestinal manifestations. *Revista do Instituto de Medicina Tropical de São Paulo*. 2003; 45(4): 197-200.
  8. Santos PH, Barros RD, Gomes KV, Nery AA, Casotti CA. Prevalence of intestinal parasitosis and associated factors among the elderly. *Revista Brasileira de Geriatria e Gerontologia*. 2017; 20(2): 244-53.
  9. Mahmoudi MR, Kazemi B, Haghighi A, Karanis P. Detection of *Acanthamoeba* and *Toxoplasma* in river water samples by molecular methods in Iran. *Iranian Journal of Parasitology*. 2015; 10(2): 250-7.
  10. Velimirovic B. Toxoplasmosis in immunosuppression and AIDS. *Infection*. 1984; 12(5): 315-7.
  11. Gallino A, Maggiorini M, Kiowski W, Martin X, Wunderli W, Schneider J, et al. Toxoplasmosis in heart transplant recipients. *European Journal of Clinical Microbiology and Infectious Diseases*. 1996; 15(5): 389-93.
  12. Wanke C, Tuazon CU, Kovacs A, Dina T, Davis DO, Barton N, et al. *Toxoplasma* encephalitis in patients with acquired immune deficiency syndrome: diagnosis and response to therapy. *The American Journal of Tropical Medicine and Hygiene*. 198; 36(3): 509-16.
  13. Hamidi F, Etemadi J, Ghabouli Mehrabani N, Mahami Oskoue M, Motavalli R, Ardalan MR. Comparison of *Toxoplasma gondii* seropositivity in hemodialysis and peritoneal dialysis patients. *Journal of Coastal Life Medicine*. 2015; 3(8): 621-2.
  14. Luft BJ, Remington JS. Clinical infectious diseases: an official publication of the infectious diseases society of America. *Toxoplasmic Encephalitis in AIDS*. 1992; 15(2): 211-22.
  15. Ferreira MS, Borges AS. Some aspects of protozoan infections in immunocompromised patients: a review. *Memorias do Instituto Oswaldo Cruz*. 2002; 97(4): 443-57.
  16. Cong W, Liu GH, Meng QF, Dong W, Qin SY, Zhang FK, et al. *Toxoplasma gondii* infection in cancer patients: prevalence, risk factors, genotypes and association with clinical diagnosis. *Cancer Letters*. 2015; 359(2): 307-13.
  17. Foroutan M, Rostami A, Majidiani H, Riahi SM, Khazaei S, Badri M, et al. A systematic review and meta-analysis of the prevalence of toxoplasmosis in hemodialysis patients in Iran. *Epidemiology and Health*. 2018; 40: 1-8.
  18. Yan C, Liang LJ, Zheng KY, Zhu XQ. Impact of environmental factors on the emergence, transmission and distribution of *Toxoplasma gondii*. *Parasites & Vectors*. 2016; 19(1): 1-7.
  19. Bora I, Dutta V, Lyngdoh WV, Khyriem AB, Durairaj E, Phukan AC. Study of intestinal parasites among the immunosuppressed patients attending a tertiary-care center in Northeast India. *International Journal of Medical Science and Public Health*. 2016; 5(5): 924-30.
  20. Naves MM, Costa-Cruz JM. High prevalence of strongyloides stercoralis infection among the elderly in Brazil. *Revista do Instituto de Medicina Tropical de São Paulo*. 2013; 55(5): 309-13.
  21. Kamani J, Mani AU, Egwu GO, Kumshe HA. Seroprevalence of human infection with *Toxoplasma gondii* and the associated risk factors, in Maiduguri, Borno state, Nigeria. *Annals of Tropical Medicine & Parasitology*. 2009; 103(4): 317-21.
  22. Tenter AM, Heckeroth AR, Weiss LM. *Toxoplasma gondii*: from animals to humans. *International Journal for Parasitology*. 2000; 30(12-13): 1217-58.
  23. Sharif M, Ajami A, Daryani A, Ziaei H, Khalilian A. Serological survey of toxoplasmosis in women referred to Medical Health Laboratory before marriage, northern Iran, 2003-2004. *International Journal of Molecular Medicine and Advance Sciences*. 2006; 2(2): 134-7.
  24. Ocak S, Duran N, Eskiocak AF, Aytac H. Anti-*Toxoplasma gondii* antibodies in hemodialysis patients receiving long-term hemodialysis therapy in Turkey. *Saudi Medical Journal*. 2005; 26(9): 1378-82.
  25. Nahnoush RK, Youssif EM, Hassanin OM. Detection of toxoplasmosis in hemodialysis Egyptian patients using serological and molecular techniques. *Australian Journal of Basic and Applied Sciences*. 2016; 10(6): 197-201.
  26. Rezavand B, Poornaki AM, Mokhtari KR, Mohammad A, Andalibian A, Abdi J. Identification and determination of the prevalence of *Toxoplasma gondii* in patients with chronic renal failure by ELISA and PCR. *Asian Pacific Journal of Tropical Disease*. 2016; 6(5): 347-9.
  27. Nissapatorn V, Leong TH, Lee R, Init I, Ibrahim J, Yen TS. Seroepidemiology of toxoplasmosis in renal patients. *Southeast Asian Journal of Tropical Medicine and Public Health*. 2011; 42(2): 237-47.
  28. Alvarado-Esquivel C, Liesenfeld O, Torres-Castorena A, Estrada-Martínez S, Urbina-Alvarez JD, Ramos-De la Rocha M, et al. Seroepidemiology of *Toxoplasma gondii* infection in patients with vision and hearing impairments, cancer, HIV, or undergoing hemodialysis in Durango, Mexico. *Journal of Parasitology*. 2010; 96(3): 505-8.
  29. Zurainee MN, Abdullah KA, Fong MY, Hoh HB, Choon J, Rahmah N. Ocular presentations and

- Toxoplasma serology. *Journal of Health and Translational Medicine*. 2017; 5(2): 98-102.
30. Perry CE, Gale SD, Erickson L, Wilson E, Nielsen B, Kauwe J, et al. Seroprevalence and serointensity of latent *Toxoplasma gondii* in a sample of elderly adults with and without Alzheimer disease. *Alzheimer Disease & Associated Disorders*. 2016; 30(2): 123-6.
31. Alvarado-Esquivel C, Liesenfeld O, Burciaga-López BD, Ramos-Nevárez A, Estrada-Martínez S, Cerrillo-Soto SM, et al. Seroepidemiology of *Toxoplasma gondii* infection in elderly people in a northern Mexican city. *Vector-Borne and Zoonotic Diseases*. 2012; 12(7): 568-74.
32. Daryani A, Sarvi S, Aarabi M, Mizani A, Ahmadpour E, Shokri A, Rahimi MT, Sharif M. Seroprevalence of *Toxoplasma gondii* in the Iranian general population: a systematic review and meta-analysis. *Acta Tropica*. 2014; 137: 185-94.
33. Saeidinia A, Tavakoli I, Naghipour Mr, Rahmati B, Ghavami Lahiji H, Salkhori O, et al. Prevalence of *Strongyloides stercoralis* and other intestinal parasites among institutionalized mentally disabled individuals in Rasht, Northern Iran. *Iranian Journal of Parasitology*. 2016; 11(4): 527-33.
34. Salehi R, Haghghi A, Stensvold CR, Kheirandish F, Azargashb E, Raeghi S, Kohansal C, Bahrami F. Prevalence and subtype identification of *Blastocystis* isolated from humans in Ahvaz, Southwestern Iran. *Gastroenterology and Hepatology from Bed to Bench*. 2017; 10(3): 235-41.
35. Rebolla MF, Silva EM, Gomes JF, Falcão AX, Rebolla MVF, Franco RMB. High prevalence of *blastocystis* spp. infection in children and staff members attending public urban schools in São Paulo State, Brazil. *Revista do Instituto de Medicina Tropical de Sao Paulo*. 2016; 58: 1-8.
36. Duda A, Kosik-Bogacka D, Lanocha-Arendarczyk N, Kołodziejczyk L, Lanocha A. The prevalence of *Blastocystis hominis* and other protozoan parasites in soldiers returning from peacekeeping missions. *The American Journal of Tropical Medicine and Hygiene*. 2015; 92(4): 805-6.
37. Hurtado-Guerrero AF, Alencar FH, Hurtado-Guerrero JC. Occurrence of enteroparasites in the elderly population of Nova Olinda do Norte, Amazonas, Brazil. *Acta Amazonica*. 2005; 35(4): 487-90.
38. Mishra N, Tripathi SM. Intestinal Parasitosis among the elderly people in northern India. *International Journal of Scientific Research*. 2016; 5(10): 40-1.
39. Oliveira MC, Silva CV, Costa-Cruz JM. Intestinal parasites and commensals among individuals from a landless camping in the rural area of Uberlândia, Minas Gerais, Brazil. *Revista do Instituto de Medicina Tropical de São Paulo*. 2003; 45(3): 173-6.
40. Sayyari AA, Imanzadeh F, Bagheri Yazdi SA, Karami H, Yaghoobi M. Prevalence of intestinal parasitic infections in the Islamic Republic of Iran. *Eastern Mediterranean Health Journal*. 2005; 11(3): 377-83.
41. Giroto KG, Grama DF, Cunha MJ, Faria ES, Limongi JE, Pinto RD, Cury MC. Prevalence and risk factors for intestinal protozoa infection in elderly residents at Long Term Residency Institutions in Southeastern Brazil. *Revista do Instituto de Medicina Tropical de São Paulo*. 2013; 55(1): 19-24.
42. Thompson RA. Giardiasis as a re-emerging infectious disease and its zoonotic potential. *International Journal for Parasitology*. 2000; 30(12-13): 1259-67.
43. Mahmoudi MR, Kazemi B, Mohammadiha A, Mirzaei A, Karanis P. Detection of *Cryptosporidium* and *Giardia* (oo) cysts by IFA, PCR and LAMP in surface water from Rasht, Iran. *Transactions of the Royal Society of Tropical Medicine and Hygiene*. 2013; 107(8): 511-7.
44. Mahmoudi MR, Nazemalhosseini-Mojarad E, Karanis P. Genotyping of *Giardia lamblia* and *Entamoeba* spp from river waters in Iran. *Parasitology Research*. 2015; 114(12): 4565-70.
45. Mahmoudi MR, Ashrafi K, Abedinzadeh H, Tahvildar-Bideruni F, Haghghi A, Bandehpour M, et al. Development of sensitive detection of *Cryptosporidium* and *Giardia* from surface water in Iran. *Iranian Journal of Parasitology*. 2011; 6(3): 43.
46. Steketee RW. Pregnancy, nutrition and parasitic diseases. *The Journal of Nutrition*. 2003; 133(5): 1661-7.
47. Fletcher SM, Stark D, Harkness J, Ellis J. Enteric protozoa in the developed world: a public health perspective. *Clinical Microbiology Reviews*. 2012; 25(3): 420-49.
48. Ahmadiara E, Hajimohammadi B. Parasitic infections in nursing homes: a permanent threat for elderly health. *Elderly Health Journal*. 2017; 3(2): 55-6.
49. Karkar A. Infection control guidelines in hemodialysis facilities. *Kidney Research and Clinical Practice*. 2018; 37(1): 1-3.