Journal of Entomology and Zoology Studies

Available online at www.entomoljournal.com

E-ISSN: 2320-7078
P-ISSN: 2349-6800
JEZS 2015; 3 (2): 42-46

Received: 02-02-2015
Accepted: 07-03-2015

Nowara E. El Ammari
Department of Microbiology and Parasitology Faculty of Medicine Benghazi University Post Box No. 18251 Benghaz, Libya.

Govindapillai A. Nair
Environmental Resources Research Centre (ERRC)
Post Box 1230 Peroorkada
Thiruvananthapuram – 695005
Kerala State, India.

Correspondence:
G. Achuthan Nair
Environmental Resources Research Center,
Post Box: 1230, Perrorkada,
Thiruvananthapuram - 695 005,
Kerala State, India.
trivandrum46@gmail.com

Critical evaluation of the intestinal Protozoan parasites among Libyan and other African residents of Al-Khoms, Libya

Nouara E. El Ammari, Govindapillai A. Nair

Abstract
Scrutiny of stools of 1250 Libyan and 1133 other African residents of Al-Khoms, Libya, during June, 2012 to May, 2013, revealed that 15.4% of the former and 25.1% of the latter contained the cysts and/or trophozoites of nine intestinal Protozoan parasites. The pathogenic among them were *Entamoeba histolytica*, *Entamoeba dispar* (recent studies confirm that *E. dispar* is a pathogen), and *Giardia lamblia*. Whether the fourth parasite *Blastocystis hominis* is a pathogen or a commensal is still not clear. The non-pathogenics were *Entamoeba coli, Endolimax nana, Chilomastix mesnili, Entamoeba hartmanni* and *Iodamoeba buetschlii.* The trophozoites of *C. mesnili* were found in the stools of Libyans only, whereas the cysts of *E. hartmanni* and *I. buetschlii* were detected in the stools of other Africans. Detailed observations were made on the single and concurrent parasitic occurrences among the two groups. Significant difference in the proportion of infection rates ($F=0.22$; $p>0.05$), and single ($F=0.000$; $p>0.05$) and concurrent ($F=0.01$; $p>0.05$) parasitic occurrences between Libyans and other Africans were not discernible. The importance of adopting preventive measures to control the spread of these parasites is stressed.

Keywords: Infection rate, intestinal parasite, Libyan and other African, pathogenic, non-pathogenic, Protozoa, single, concurrent, stool

1. Introduction
Population movement has been cited as one of the major factors spreading the parasites worldwide and the epidemiological patterns of parasitic diseases is further complicated in a developing country like Libya by the arrival of large numbers of migrant workers from the neighboring and sub-saharan African countries, leading to de-stabilizing effects on the normal pattern of disease transmission [1]. The present study was undertaken during the period June, 2012 to May, 2013, on 2383 Libyan and other African residents of Al-Khoms, Libya, and their stools were examined to detect the occurrence of cysts and/or trophozoites of intestinal protozoan parasites. The study gives an insight on the occurrence of intestinal protozoan parasites among them and the importance of taking preventive and precautionary measures to control them. Earlier studies on the parasitic infections in Libya were mainly confined on school children [2, 3, 4]. Later, detailed studies on the intestinal protozoan and helminth parasites among the Libyan and alien residents of Benghazi, Libya, were published [3, 5, 6].

2. Material and methods
2.1. Place of study
The study was conducted in Al-Khoms (32º38'54"N, 14º15'42"E) (Fig.1), a city on the Mediterranean coast of Libya in the north-west about 97 km south-east of the capital city Tripoli. It is a tourist destination.

2.2. Details about nationals examined
Al-Khoms attracts large number of migrant workers from the neighbouring north-African and sub-Saharan countries for employment. Some engage in business also. The local Libyan populations are Arabs and the other African nationals residing in Al-Khoms are both Arabs and non-Arabs. The different nationalities (age: 15-55 years) whose stools were examined Were divided into 1) Libyans, who were settled for generations in Al-Khoms, and 2) other Africans, who were residing in Al-Khoms for the past 3 to 5 years. Out of the total 2383 nationals randomly selected for the study, 1250 (800 males and 450 females) were Libyans, and 1133 (890 males and 243 females) were other Africans (Egypt, Sudan, Chad, Morocco,

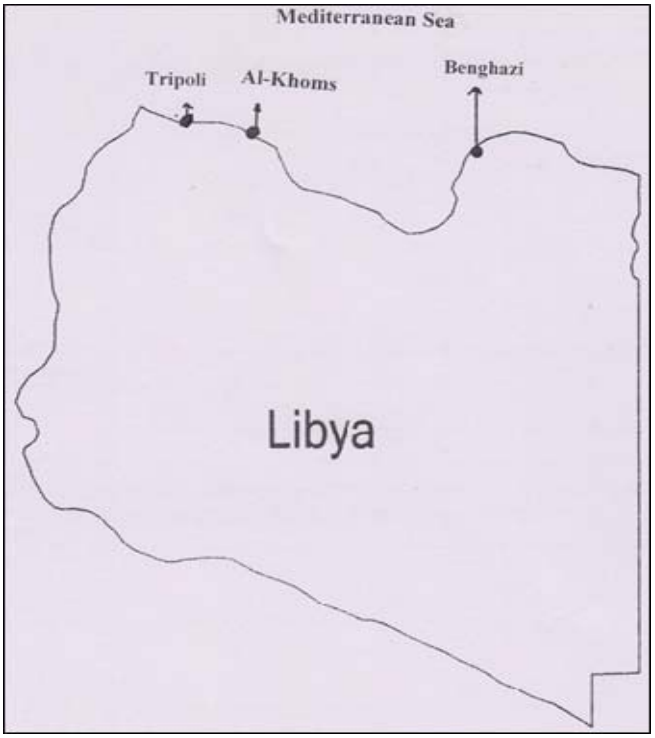

Fig 1: Map of Libya showing the location of Al-Khoms

Tunisia, Algeria, Niger, Mauritania, Ghana, Ethiopia, Nigeria). All these nationals were required by the Municipal authorities of Al-Khoms to undergo a complete medical check-up for different purposes such as joining for new jobs, renewal of job contracts, issuing residential visas, and for obtaining licenses for trade and commerce. Some gave their stools for routine check-ups. The stools wee procured from the Central Medical Laboratory. Al-Khoms. The stools of referred sick cases from different health centres were not included in the study.

2.3. Methodology of stool examination

Fresh stools, deposited by different nationals, were collected twice a week in clean, numbered plastic containers. Personal details including name, sex, age and nationality were recorded for each sample. The consistency of the sample was checked and observations were made on the occurrence or otherwise of blood and mucus in the stools. They were later homogeneously mixed and examined on the same day of collection by direct smear examination using both normal saline solution and Lugol's iodine [7], and by Zinc sulphate [8]. The cysts and trophozoites of parasites found in the stools were identified using keys and descriptions [9].

2.4. Statistical analysis

The ANOVA [10] were done to determine whether there existed a significant difference or not on the proportions of infection rates and on the single and concurrent parasitic infections between Libyan and other African nationals residing in Al-Khoms, Libya.

3. Results

3.1. Intestinal protozoan parasites identified in the study

The cysts and/or trophozoites of protozoan parasites isolated from the stools of Libyans and other Africans were 1) *Entamoeba histolytica* (cyst), 2) *Entamoeba dispar* (cyst), *Giardia lamblia* (cyst and trophozoite), *Entamoeba coli* (cyst), *Blastocystis hominis* (vacuolated cyst), *Endolimax nana* (cyst), *Chilomastix mesnili* (trophozoite), *Entamoeba hartmanni* (cyst) and *Iodamoeba buetschlii* (cyst). Of these *E. histolytica* and G. *lamblia* wee pathogenetics causing the diseases amoebiasis and giardiasis respectively. Recent findings [11, 12] indicate that *E. dispar* may also be a pathogen. The remaining parasites except *B. hominis* were either commensal or non-pathogenic. Whether *B. hominis* is a pathogen or commensal is still not clear. The cysts of *E. histolytica* and *E. dispar* were grouped together as *E. histolytica/E. disar* in the present study, since it was difficult to differentiate between the cysts of these parasites.

3.2. Infection rates

Infection rates of Libyan and other African nationals living in Al-Khoms are presented in Table 1, and the number and percentages of these groups infected with different protozoan parasites, out of the total examined in each group, are given in Table 2. The trophozoites and/or cysts of protozoan parasites except those of *E. hartmanni* and *I. buetschlii* were found in the stools of 192 (15.4%) Libyans, and all the parasites except those of *C. mesnili* were detected in the stools of 284 (25.1%) other Africans. The differences in the proportions of infection rates were not found to be significant ($F=0.22$; $p>0.05$) between Libyans and other Africans.

The cysts of *E. histolytica/E. dispar* were recorded in 9.8% Libyans and 14.7% other Africans. 7% and 3.5% Libyans and other Africans respectively were found infected with *G. lamblia*. On the other hand, the prevalence rate of *E. coli* was higher in other Africans (4.1%) when compared with the rate among Libyans (2.6%). The vacuolated cysts of *B. hominis* in the stools of Libyans and other Africans were 0.2% and 0.9% respectively. The cysts and trophozoites of *Endolimax nana* and *Chilomastix mesnili* were located in the stools of 0.9% and 0.2% Libyans respectively whereas the former were found in 1.3% and the latter were absent in the stools of other Africans. The cysts of *E. hartmanni* (0.3%) and *I. buetschlii* (0.3%) were detected in the stools of other Africans only.

3.3. Single and concurrent parasitic infections

Single and concurrent infections among Libyan and other African residents of Al-Khoms by intestinal protozoan parasites are presented in Table 3. Regarding single parasitic infection, 53% of the schools of Libyan and 44.1% of the same of other Africans had the cysts of *E. histolytica/E. dispar*. This was followed by the next higher percentages of Libyans (4.2%) recording the cysts and/or trophozoites of *G. lamblia*, and the cysts of *E. coli* (3.6%) in their stools. The corresponding percentages of *G. lamblia* and *E. coli* among other Africans were 7% and 8.8% respectively. The vacuolated cysts of *B. hominis* were detected in the stools of 0.5% Libyans and 1.8% other Africans. The cysts of *E. nana* were found in the stools of 2.2% Libyans and 2.8% other Africans.

Table 1: Infection rates of Libyans and other African nationals residing in Al-Khoms, Libya (2012-2013) having cysts and/or trophozoites of intestinal protozoan parasites in their stools

Country	Number tested	Number infected	%
Libya	1250	192	15.4
Other Africans			
Egypt	420	98	23.3
Sudan	312	70	22.4
Chad	80	30	37.5
Morocco	88	20	22.7
Tunisia	76	19	25
Ghana	53	19	35.8
Algeria	40	12	30
Others*	64	16	25
	1133	**284**	**25.1**

* Number of subjects tested of each of other nationalities (Niger, Mauritania, Mali, Ethiopia, Nigeria) <20

Table 2: Infection rates with various intestinal protozoan species (cysts and/or trophozoites) among Libyan and other African residents of Al-Khoms, Libya (2012-2013)

Parasite Species	Libyan (1250)		Other African (1133)	
	Number infected	%	Number infected	%
Entamoeba histolytica/ Entamoeba dispar	122	9.8	167	14.7
Giardia lamblia	21	7	40	3.5
Entamoeba coli	32	2.6	46	4.1
Blastocystis hominis	3	0.2	10	0.9
Endolimax nana	12	0.9	15	1.3
Chilomastix mesnili	2	0.2	0	0.0
Entamoeba hartmanni	0	0.0	3	0.3
Iodamoeba buetschlii	0	0.0	3	0.3
All	192	15.4	284	25.1

Coming to double parasitic infections, 8.3% Libyans and 6.3% other Africans were found infected with the cysts of *E. coli* + the cysts/trophozoites of *G. lamblia* in their stools. This was followed by 7.3% and 2.5% of Libyans and other Africans respectively infected with the cysts of *E. histolytic*/ *E. dispar* + the cysts of *E. coli* in their stools. The cysts and/or trophozoites of *G. lamblia* were detected in the stools of 5.2% Libyans and 7.0% other Africans. 2.2% Libyans and 2.5% other Africans were found to have the cysts of *E. histolytica/E. dispar* + vacuolated cysts of *B. hominis* in their stools. The cysts of *E. coli* + *E. nana* were found in the stools of 4.2% and 6.3% Libyans and other Africans respectively.

Regarding triple parasitic infections, the cysts of *E. histolytica/E. dispar* + cysts of *E. coli* + the trophozoites of *C. mesnili*; the cysts of *E. historytical/E. dispar* + the cysts of *E. coli* + the cysts of *E. nana* were detected in the stools of 3.1% and 6.2% Libyans respectively. Such a combination was not found in the stools of other Africans. On the other hand, 1.1%, 1.1% and 8.7% of the stools of other Africans contained the 1) the cysts of *E. histolytica*/*E. dispar* + the cysts of *E. nana* + the cysts of *E. hartmanni*; 2) the cysts of *E. histolytica*/*E. dispar* + the cysts and/or trophozoites of *G. lamblia* + the cysts of *I. buetschlii*, 3) the cysts of *E. nana* + the cysts of *E. coli* and the cysts and/or trophozoites of *G. lamblia* in their stools. Similar combinations were not found in the stools of Libyans.

Significant differences of the single ($F=0.000$; $p>0.05$), and concurrent ($F=0.01$; $p>0.05$) parasitic infections between Libyans and other Africans were not discernible.

4. Discussion

The cysts of *E. histolytica*/*E. dispar* showed widespread prevalence in the stools of Libyans and other Africans residing in Al-khoms, Libya. Intestinal Protozoa are the etiological agents of several widespread diseases, the most common of which are caused by *E. histolytica* [13]. Intestinal amoebiasis caused by *E. histolytica* is the third principal parasitic disease responsible for mortality in the world. This protozoan parasite infects approximately 180 million individuals throughout the world among which 40 to 100 thousand die each year WHO. [14]. Amoebiasis is a cosmopolitan infection transmitted by fecal-oral route, food and drink and its greatest impact is in Africa and in Asia. In Africa, Egypt, Morocco, South Africa, Cote d'lvoire and countries located between 10ºN

Table 3: Percentages of positive cases of Liban (n=192) and other African nationals (n=284) residing in Al-Khoms (2012-2013) having single or concurrent parasitic occurrences of the cysts and/or trophozoites of intestinal protozoan parasites in their stools.

Sl. No.	Parasites	% positive cases in	
		Libyans	Other Africans
	Single parasitic occurrence		
1	*Entamoeba histolytica/Entamoeba dispar*	53	44.1
2	*Giardia lamblia*	4.2	7.0
3	*Entamoeba coli*	3.6	8.8
4	*Blastocystis hominis*	0.5	1.8
5	*Endolimax nana*	2.2	2.8
	Concurrent parasitic occurrence		
1	*E. histolytica/E. dispar + E. coli*	7.3	2.5
2	*E. histolytica/E. dispar + B. hominis*	2.2	2.5
3	*E. histolytica/E. dispar + G. camblia*	5.2	7.0
4	*E. coli + E. nana*	4.2	6.3
5	*E. coli + G. lamblia*	8.3	6.3
6	*E. histolytica/E. dispar + E. coli + C. mesnili*	3.1	0.0
7	*E. histolytica/E. dispar + E. coli + E. nana*	6.2	0.0
8	*E. histolytica/E. dispar + E. nana + B. hartmanni*	0.0	1.1
9	*E. histolytica/E. dispar + G. lamblia + I. buetschlii*	0.0	1.1
10	*E. nana + E. coli + G. lamblia*	0.0	8.7

And 10 ºS are severely affected [14, 15]. This categorization includes Libya also [1, 5, 6]. Moderate to high prevalence of this parasite has also been reported in populations residing in sub-Saharan and Arabian countries [16, 17].

Moderate percentages of Libyans and other Africans of Al-Khoms had the cysts/trophozoites of *G. lamblia* in their stools. Giardiasis, caused by *G. lamblia* infects approximately 200 million individuals throughout the world, is a frequent cause of diarrhea in children, and can have negative impact on growth and development [14]. Giardiasis is reported earlier among the school children of Benghazi [2, 3]. The prevalence of giardiasis and mixed giardiasis/amoebiasis infections were significantly higher in malaria positive subjects than in the malaria-free group [18]. *G. lamblia* is the most prevalent intestinal parasite

among school children in different parts of Ghana [19]. With the advent of AIDS, there was speculation that *G. lamblia* may be an important pathogen of this group, but clinical findings to date do not seem to confirm this possibility [20]. Giardiasis is also reported among Jordanian [21], Sand, Arabian [22] and Palestinian [17] populations. High prevalence of giardiasis in the Libyan, other Arab and non-Arab populations living in Benghazi was reported [5]. Also moderate to high prevalence of the same were detected among the non-Libyan African and non-African residents of Benghazi, Libya [1].

It was believed that *E. dispar* is a non-pathogen unable to produce significant experimental lesions. This scenario changed when *E. dispar* strains were isolated from symptomatic patients in Brazil. These strains were able to produce liver and intestinal lesions that are occasionally indistinguishable from those produced by *E. histolytica*. These and other finding have revived the possibility that *E. dispar* can produce lesions in humans [11]. Recent experimental studies also confirm that *E. dispar* can be considered as a pathogen [12].

A low prevalence of the vacuolated cysts of *B. hominis* were found in the stools of both Libyan and other African residents of Al-Khoms. *B. hominis* is found in the stools of both healthy people and in the stools of those who have diarrhea, abdominal pain and other gastrointestinal problem [23]. Infection with *B. hominis* is called blastocystosis. The most critical question concerning *B. hominis* is whether it is a pathogen or a commensal. Several reports suggest that *B. hominis* may be an opportunistic infection in immune suppressed patients with AIDS [24, 25], but whether as a commensal or as a pathogen remains to be determined.

E. coli, E. nana, E. hartmanni, I. buetschlii and *C. mesnili* are generally considered non-pathogenics and found in the lumen of the intestinal tract. They are not found in the cells that line the intestine and do not spread to other parts of the body. Both the cysts and the trophozoites of these species are passed in stools and considered diagnostic. Cysts are typically found in formed stools whereas trophozoites are usually found in diarrheal stools. Colonisation of the non-pathogenic amoeba occurs after ingestion of mature cysts in fecally contaminated food, water or fomites. Encystations occurs in small intestine and trophozoites are released, which migrate to the large intestine. The trophozoites multiply by binary fission and produce cysts and both stages are passed in the feces. The cysts can survive days to weeks in the external environment and are responsible for transmission. Trophozoites passed in the stools are rapidly destroyed once outside the body, and if ingested would not survive exposure to the gastric environment [26]. Moderate percentages of Libyans and other Africans had the cysts of *E. coli* and *E. nana* in their stools. High prevalence of *E. coli* was reported earlier among the school children of Benghazi [3]. The recognition of non-pathogenic protozoan parasites is generally accepted as a useful epidemiological indication of the level of fecal contamination [1, 5, 22]. The non-pathogenic forms recorded in the present study are cosmopolitan in distribution and are harmless except for creating intestinal disturbances, if found in large numbers.

5. Conclusion

High prevalence of *E. histolytica, E. dispar* (which is now considered as a pathogen) and *G. lamblia,* and their combinations, the low prevalence of *B. hominis* and their combinations with pathotenic forms in Libyan and other African residents of Al-Khoms, are a matter of concern. The gravity of the situation increases further after the recent findings that *G. lamblia* and *B. hominis* are important and opportunistic parasites related with malaria and AIDS. Fortunately malaria is not prevalent in Libya, but the possibility of carrying malarian parasites by other Africans is high. The transmission of some of the parasites found in the present study from the alien African population to the indigenous Libyans and vice versa cannot be ruled out unless proper preventive and precautionary measures are implemented to check the spreading of these parasites. These include effective sanitation to prevent water and food contamination, proper sewage, adequate handling and treatment of water supplies, and health education. Stringent parasitological surveillance of immigrant population may be of value in checking the transmission of diseases.

6. Acknowledgements

Thanks are due to M/s. Classic Copiers, Thiruvananthapuram, Kerala, India for the DTP printout of the Ms.

7. References

1. Nowara EE, Nair GA. Intestinal protozoan parasites among Libyan, non-Libyan African and non-African residents of Benghazi, Libya. Revista Iberica de Parasitologia 2005; 65(1-4):15-20.
2. Dar FK, Khously-ElSI, Boulaqi-ElHA, Murir R, Maghrabi-ElS. Intestinal parasites in Benghazi school children. Garyounis Medical Journal 1979; 2:3-7.
3. Boulaqi-ElMA, Dar FK, Medini MS. Prevalence of intestinal parasites in primary school children in Behghazi city. Journal of Egyption Society of Parasitology 1980; 10:77-82.
4. Buni-ElAA, Khan AH. Intestinal protozoan infection in Benghazi. Sebah Medical Journal 1998; 1:106-108.
5. Ammari-ElNE, Nair GA, Kassem HH. Intestinal protozoan and helminth parasites among Libyans, non-Libyan Arabs and non-Arabs living in Benghazi, Libya. Jordan Journal of Applied Science 2004; 6(2):72-81.
6. Ammari- ElNE, Nair GA. Occurrence and prevalence of intestinal protozoan parasites in male and female Libyan nationals residing in Benghazi, Libya Revista Iberica de Parasitologia 2003; 63(3-4):47-53.
7. Markell ED, John DT, Krotoski WA. Medicasl Parasitology. WB Saunders, Philadelphia, USA, 1999.
8. Zeilbig EA. Clinical Parasitology. WB Saunders, Philadelphia, USA, 1997.
9. Neva FA, Brown HW. Basic Clinical Parasitology. Appleton and Lange, Connectient, USA, 1994.
10. Grimm LG. Statistical Applications for the Behavioral Sciences. John Wiley and Sons, New York, USA, 1993.
11. Oliveira FMS, Neumann E, Gomes MA, Caliari MV. *Entamoeba dispar*: could it be pathogenic. Tropical Parasitology 2015; 5(1):9-14.
12. Graffeo RG, Archibusacci CM, Soldini S, Romano L, Masucci L. *Entamoeba dispar*: A rare case of enteritis in a patient living in non-endemic area. Case Report: Gastrointestinal Medicine published, 2014. Online doi: 70.1155/2014/498058.
13. Schnunis GA, Lopez Antumano FJ. Importance of parasites. In: Parasitology. Arnold, London, UK, 1998.
14. WHO. Intestinal Protozoan and Helminth Infections. WHO Technical Report, Series, 1981, 666.
15. Stauffer W, Abd-Alla M, Ravdin JL. Prevalence and incidence of *Entamoeba histolytica* infection in South Africa and Egypt. Archiv Medical Research 2006; 37(2):266-269.
16. Gatti S, Mahdi R, Bruno A, Cevini C, Seaglia M. A

survey of amoebic infection in the Wonji area of central Ethiopia. Annals Tropical Medicine 1998; 92:173-179.
17. Shubair ME, Yassin MM, AL-Hindi AL, Al-Wahaidi AA, Jadallah SJ, Abushaban N. Intestinal parasites in relation to hemoglobin level and nutritional status of school children in Gaza. Journal of Egyptian Society of Parasitology 2000; 30:365-375.
18. Nwanguma B, Alumanah E. Concurrent giardiasis and amoebiasis infections in Nigerian children diagnosed with *plasmodium palciparum* malaria: prevalence and patho-physiological implications. Internet Journal of Tropical Medicine 2008, 6(1). Cited. http://ispub.com/IJTM/6/1/11038.
19. Nkrumah B, Nguah SB. *Giardia lamblia*: a major parasitic cause of childhood diarrhea in patients attending and district hospital in Ghana. Parasites and Vectors; 4: doi. 10. 1186/1756-3305-4-163.
20. Meyer EA. Giardiasis. Elsevier, Amstardam, Holland, 1990.
21. Morsy TA, El-Maridi NA. Incidence of parasitic infection in Baqaa. Journal of Egyptian Society of Parasitology, 8:347-351.
22. Hammouda NA, Lehshtein AK, Abdel Fattah MM, Wasfi AS, Omar ER, Higazi NA. parasitic infections and nutritional status of school children in the western region of Saudi Arabia. Journal of Egyptian Society of Parasitology 1986; 16:675-682.
23. Telalabasic S, Pikula Z, Kipidzic M. *Blastocystis hominis* may be a potential cause of intestinal disease. Scandanavian Journal of Infections Diseases 1991; 23:389-390.
24. Garavelli PL, Siaglione L. *Blastocystis hominis* infection in AIDS and correlated pathologies. Minnera Medica 1990; 81:91-92.
25. Vallano A, Pigran C, Hermandez A, Garalda J. *Blastocystis hominis* in a HIV positive homosexual patient. Revue Clinical Espana 1991; 188:110-111.
26. Parasites. Non-pathogenic (harmless) intestinal Protozoa. Published by Centre for Disease Control and Prevention [www.ede.gov/parasites/nonpathprotozoa] 2012.