Study on the Role of Pet Animals for *Helicobacter pylori* Transmission

Mohamed Abdel-Raouf[1], YousefAbdel-Gleel[2], Ayman Enab[3]

[1]Gastro - Enterology Center, Mansoura University, Mansoura, Egypt
[2]Microbiology Department, Faculty of Vet. Medicine, Zagazig University, Egypt
[3]Microbiology Department, Faculty of Vet.Medicine, Zagazig University, Egypt
mohamed.raouf999@yahoo.com

Abstract: *Helicobacter pylori* infection is one of the most common bacterial infections in humans. Although *H. pylori* may be detected in the stomach of approximately half of the world's population, the mechanisms of transmission of the microorganism from person to person are not yet clear. Transmission of *H. pylori* could occur through, fecal-oral, and oral-oral routes, and through food and water**.** *Helicobacter* colonizes the stomachs and intestines of humans and several animal species, such as cats, dogs. Prevalence in healthy a symptomatic persons and in apparently healthy dogs and cats. *H. pylori* might have jumped quite recently from animal hosts to people. Because it has been possible to transfer from humans to animals it's reasonable to suppose that animals might have been the original source of the bacterium**.** The aim of this work is to investigate the role of pet animals in *Helicobacter pylori* transmission and, to study the antibiotic susceptibility of *H. pylori* isolate. Recent reports suggest that the overall prevalence of *H. pylori* in stool, saliva and, stomach juice samples of examined dogs are 41.4 %, 42.9%, and 50%, respectively. Although the incidence of *H. pylori* in stool, saliva, and stomach juice samples of examined cats are 30 %, 42.9% and 20 %, respectively. Moreover, we discussed the incidence of *H. Pylori* in stool and saliva samples of healthy persons related to dogs ware 23.9% and 8.7%, respectively, but in diseased persons ware 64.3 %and 42.9 %, respectively. Also, the incidence of *H. Pylori* in gastric juice samples of diseased men and women's related to dogs with an incidence were 60 % and 66.7 %, respectively. Whole, the incidence of *H. pylori* in stool and saliva samples of healthy persons related to cats were 40.9 % and 45.5 %, respectively, however diseased persons were 75 % and 62.5 %, respectively. The prevalence of *H. pylori* in gastric juice samples of diseased men and women's related to cats were 66.7 % and 80 %, respectively. *H. pylori* isolates sensitive to cefotaxime sodium**,** ceftazidime**,** ceftraxone**,** cefuroxime sodium**,** ciprofloxacin**,** levofloxacin**,** meropenem**,** nitrofurantion and ofloxacin. Thus,*H. pylori* can be isolated from feces, saliva and stomach juice of dogs, cats and human by microbiological methods without need of Endoscopy. Pet animals may be a good reservoirs for the transmission of *H. pylori* between domestic pets and their owners.

[Mohamed Abdel-Raouf, Yousef Abdel-Gleel, Ayman Enab. **Study on the Role of Pet Animals for *Helicobacter pylori* Transmission.** *J Am Sci* 2014;10(8s):20-28]. (ISSN: 1545-1003). http://www.jofamericanscience.org. 2

Keywords: Pet animals;*Helicobacter pylori*; Transmission; Incidence

1- Introduction.

H. pylori is Gram negative, have a spiral or helical shape, and motile by several (has 4-6 sheathed flagella attached to one pole which allow high motility) and a smooth surface (Malfertheiner and Pieramico, 1992 and Forbes *et al.,* 2002). Hazell *et al.* (1987) found that *H. pylori* is strongly urease, oxidase, and catalase positive.*H. pylori* requires special environmental and cultural conditions for its growth as it is fastidious, microaerophilic conditions (10% CO_2 and 5% O_2), growth is best on moist freshly prepared heated (chocolate) blood agar, translucent colonies appear after 3 to 7 days incubation, but when it growth in supplemented broth, like Brucella Broth with fetal calf serum 1-10% (Goodwin *et al.,* 1985).

H. pylori colonize the surface of the mucosa, especially of the antrum, and the overlaying layer of mucus and it extends into gastric glands (Mobley, 1997). It has developed several characteristics (virulence factors) to survive in the interior surface of the stomach beneath a thick mucus layer(Jerris, 1995). *H. pylori* infection is one of the major discoveries in gastroenterology within the past twenty years. Human and animal gastric mucosa is the natural ecologic niche of *H. pylori*.

However, unlike the commensal microorganisms that inhabit mucosal surfaces, *H. pylori* is capable of causing inflammation and disease at the site of infection. The inflammatory process does not lead to clearance of this infection.Possibly this pathogen has adapted to colonization of inflamed mucosal surfaces, making inflammation a prerequisite to prolonged colonization.

The majority of infected individuals do not suffer from associated gastrointestinal disease, but a proportion of infected persons develop acute gastritis, peptic ulcer disease, mucosa-associated lymphoid tissue lymphoma (MALT), or gastric adenocarcinoma.

H. pylori infection is considered a major cause of these conditions(Suerbaum and Michetti, 2002). Although peptic ulcer disease is the most studied disease related to *H. pylori* infection, this bacterium is seemingly involved in the pathogenesis of several extra gastric diseases, such as gastro esophageal reflux disease, iron deficiency anemia, skin disease, and rheumatologic conditions (Rothenbacher and Brenner, 2003**).**

H. pylori colonized in GIT of gnotobiotic dogs orally challenge exposed with 3 x 10^8 organisms at 7 days of age. It had colonized in all parts of the stomachs of live dogs: cardio, funds, antrum, and pyloric antrum. *H. pylori* in dogs was similar to that in humans and is transmissible by direct contact (Radin *et al.,* 1990). Lavelle *et al.* (1994) suggested that dogs, cats, and pigs may have been the source of infection to humans.

Isolation of *H. pylori* from cats which is confirmed by morphological and biochemical evaluations, fatty acid analysis, and 16 S rRNA sequence analysis. In most animals, *H. pylori* was present in close proximity to mucosal epithelial cells or in mucus layers of the glandular or surface epithelium (Handt *et al.,* 1994).Isolation of *H. pylori* from 50% of saliva and 90% of gastric juice of domestic cat has also been suggesting that the organism could be a zoonotic pathogen with transmission from infected animals who have close human contact, and it can promote gastritis when introduced into specific-pathogen-free cats which demonstrated multifocal gastritis in cats experimentally infected with *H. pylori.* (Fox, 1995, and Handt *et al.,* 1995**).** Domestic cats is a common naturally infected host with *H. pylori* which readily colonizes the cat stomach and can subsequently be detected in feline saliva and feces (Fox *et al.,* 1995).*H. pylori* has been cultured, PCR -amplified from the feces, salivary secretions, gastric fluids and dental plaque of naturally- infected cats(Christopher, 2004*)*.Close contact in human and animals seems to enhance the transmission of *H. pylori*, which is suspected to be transmitted by oral-oral, fecal-oral or gastric-oral routes, which are evidenced by isolation of *H. pylori* from saliva and feces(Taylor and Blaser*,* 1991*)*.

Direct urease test, brush cytology, histopathology, electron microscopy, PCR and other molecular analyses of gastric biopsies, gastric juice, or culture isolates can be used to diagnosis and identify of *H. pylori* (De Boer, 1997). The most specific way to diagnosis of *H. pylori* is microbiological culture, it is essential to identify the bacteria (Hachem *et al.,* 1995, Lopez –Brea *et al.,* 1997; Topley and Wilson, 1998).

Increasing prevalence of drug-resistant for *H. pylori* infections necessitate susceptibility testing methods such as micro broth dilution, disk diffusion, and agar dilution have been used to assess antimicrobial resistance in *H. pylori*. The relationship among previous antimicrobial use, antimicrobial resistance, and the treatment outcomes for *H. pylori* infections (Toracchio and Marzio. 2003; Nicola *et al.,* 2004; Boyanova *et al.,* 2006)

2- Material and Methods.

Samples:

A total number of 70 stool, 70 saliva and 40 stomach juice samples from dogs,and, 70 stool, 63 saliva and 50 stomach juice samples from cats were randomly collected from different breeds hosted in various localities. Such samples were gathered with an empty stomach in the early morning.

Table 1: Samples collected from both domestic and stray dogs and cats

Dogs				Cats		
	Domestic	Stray	Total	Domestic	Stray	Total
Stool	53	17	70	52	18	70
Saliva	53	17	70	45	18	63
Stomach juice	30	10	40	35	15	50
Total	136	44	180	132	51	183

Table 2: Number and nature of collected samples from persons related to dogs(M: male and F: Female).

Sample	**Persons related Dogs**						
	Healthy		**Diseased**		**Total**		**Total**
	M	**F**	**M**	**F**	**M**	**F**	
Stool	**38**	**6**	**10**	**6**	**48**	**12**	**60**
Saliva	**38**	**6**	**10**	**6**	**48**	**12**	**60**
Stomach juice	**0**	**0**	**10**	**6**	**10**	**6**	**16**
Total	**76**	**12**	**30**	**18**	**106**	**30**	**136**

Table 3:Number and nature of collected samples from persons related to cats(M: male and F: Female)

Samples	Persons related cats						
	Healthy		Diseased		Total		Total
	M	F	M	F	M	F	
Stool	16	28	6	10	22	38	60
Saliva	16	28	6	10	22	38	60
Stomach juice	-	-	6	10	6	10	16
Total	32	56	18	30	50	86	136

A total number of 60 stool, 60 saliva and 16 stomach juice samples were collected from each person's related to the examined dogs and cats in various localities (Tables 2 and 3). Such samples were gathered from persons with an empty stomach in the early morning.Persons related whom suffered from gastrointestinal disorders were subjected to Endoscopy unit in Gastroenterology center, Mansoura University (Endoscopy Olympus fibro optic GIF type xq 20-24/34/s).The samples of gastric juice were collected by endoscopy under strict a septic precautions from each case.

All examined stool samples were collected in sterile plastic bags. A part of such samples were directly cultured into sterile cork screw plastic tube contained 7-10 ml Selenite-F –Broth as enriched transport medium to support the growth of the bacterium. After appearance of signs of action of Xylaject, lowering of the head, drooping of the lower lip, salivation, protrusion of the tongue saliva were collected from dogs and cats by sterile cotton swabs.Such samples were added to 2-3 ml physiological saline solution(as transport medium). Stomach juice samples were collected from dogs and cats, respectively with empty stomach on the morning under strict a septic precautions after intramuscular injection of 1-1.5 ml Xylaject for such cat and 2-3 ml for such dog to help in saliva collection and to induce vomition within 5 minutes. Each sample was added to 2-3 ml physiological saline solution (as transport medium) in sterile cork screw plastic tube.

Collection of saliva samples from persons in sterile plastic bags. Gastric juice samples were picked up from persons whom suffered from gastrointestinal disorders by using Esophagogastroduodenoscopy under strict a septic precautions. Each sample was treated with 2-3 ml physiological saline solution in sterile cork screw plastic tube.

Sample was shacked well and then inoculated on Brain Heart Infusion Chocolate Agar plates (10% blood) (CM 331, Oxiod, England) supplemented with *H. pylori* selective supplement (Dent, SR 0147 E Oxoid Limited, Basingstoke, Hampshire,England), incubated in CO_2 incubator (microaerophilic condition of 5% O_2,10% CO_2, 85% N_2 and 99% relative humidity) at 37^oC. Cultures were inspected following 3,5 and 7 days incubation.

The growing colonies subjected to criteria of both colonial morphology, Gram stain and biochemical identification as *H. pylori* on the basis of the production of urease, catalase and oxidase enzymes. *H. pylori* susceptibility test was performed with disc diffusion technique.

Statistical Analysis.

Done with aid of statically reference:Klein. D. (1996) "Logistic Regression "Springer-verlag, New York. A confidence interval (CI) is an indicator of the measurement's precision. It is also an indicator of how stable the estimate is, which is the measure of how close the measurement will be to the original estimate if the experiment is reported. Follow the steps below to calculate the confidence interval for your data.

3- Results.

The microorganism was identified as *H. pylori by* the recommended methods, on the basis of colony morphology (small, whitish gray translucent, flat colonies), Negative Gram's stain, and the positive production of urease, catalase and oxidase enzymes.

The incidence of *H. pylori* in dogs and their related persons.

A total of 24,24 and 16 stool, saliva and stomach juice samples from domestic dogs were Positive of *H. pylori* infection, with incidence 45.3%, 45.3% and 53.3%, respectively. A total 5,6 and 3 of stool, saliva, and stomach juice samples from stray dogs were positive with incidence 29.4%, 35.3%, and 30.3%, respectively.

A total of 11 stool samples from healthy persons related dogs (10 males and one females) were positive of *H. pylori*, with incidence 23.9 % (26.3% males and 12.5% females). Nine stool samples from related persons with gastrointestinal disorders (6 males and 3 females) were positive of *H. pylori* with 64.3 % (60% males and 75% females).

A total of 4 saliva samples from healthy persons related dogs (3 males and one females) were positive of *H. pylori*, with incidence 8.7% (7.9% males and 12.5% females). Six saliva samples (5 males and one females) from related persons with gastrointestinal disorders were positive of *H. pylori* with incidence 42.9% (50% males and 25% females).

A total of 10 gastric juice samples (6 males and 4 females) from persons related dogs with gastrointestinal disorders were positive of *H. pylori*, with incidence 62.5% (60% male and 66.7% female) (Table 4).

The incidence of *H. pylori* in cats and their related persons.

A total of 16, 20 and 8 stool, saliva, and stomach juice samples from domestic cats were positive of *H.pylori* with incidence 30.8%, 44.4% and 22.9%, respectively.A total of 5,7, and 2 stool, saliva and stomach juice samples from stray cats were positive of *H.pylori* with incidence 27.8%, 38.9%,and 13.3%, respectively.

A total of 18 stool samples from healthy persons related cats (8 males and 10 females)were positive of *H.pylori* with incidence 40.9 %samples (50% male and 35.7% female).Twelve stool samples from related persons with gastrointestinal disorders (4 males and 8 females) were positive of *H. pylori* with incidence 75 % (66.% males and 75% females).

A total of 20 saliva samples from healthy persons related cats (6 males, 14 females) were positive of *H.pylori* infection with incidence 45.5% (37.5% males and 50% females).Total 10 samples (3 males and 7 females) from related persons with gastrointestinal disorders were positive of *H. pylori* with incidence 62.5% (50% males and 70% females).

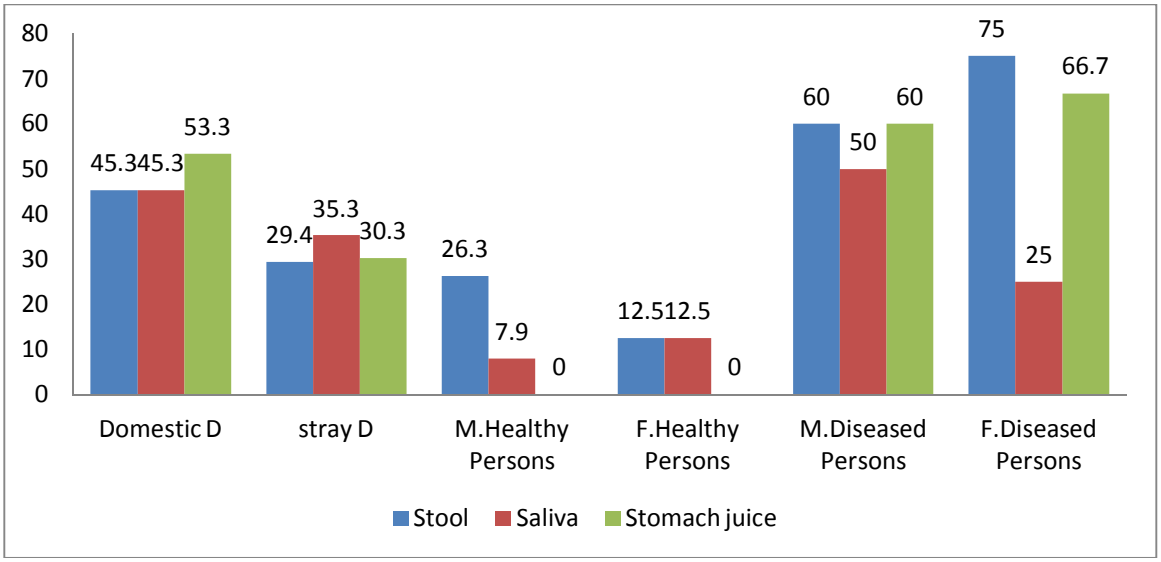
A total number of 12 gastric juice samples (4 males and 8 females) from related persons with gastrointestinal disorders were positive of *H. pylori* with incidence 75% (66.7% males and 80% females).

Sensitivity test.

After incubation the degree of sensitivity was determined by measuring the easily visible and clear zone of inhibition of growth produced by diffusion of the antimicrobial agent from the discs into the surrounding medium. The results were interoperated according to Nahar *et al.* (2004) as shown in Fig.3.

Table 4:The incidence of *H. pylori* in dogs and their related persons. (M: male and F: Female)

Sample	Dogs						Healthy Persons						Diseased Persons					
	Domestic			Stray			M			F			M			F		
	No.	P.	%	N0.	P.	%	No.	P.	%	No.	P	%	No.	P	%	No.	P	%
Stool	53	24	45.3	17	5	29.4	38	10	26.3	8	1	12.5	10	6	60	4	3	75
Saliva	53	24	45.3	17	6	35.3	38	3	7.9	8	1	12.5	10	5	50	4	1	25
Stomach juice	30	16	53.3	10	3	30.3	-	-	-	-	-	-	10	6	60	6	4	66.7
Total	76			44			76			16			30			16		

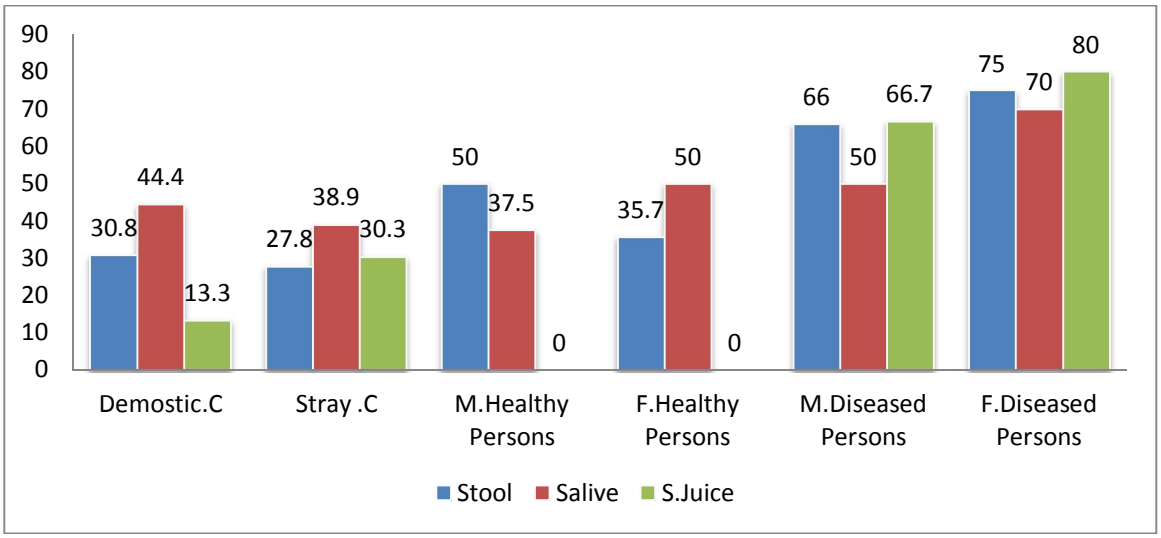


D: Dogs; M: Male; F: Female

Fig.(1):The incidence of *H. pylori* in dogs and their related persons

Table 5: The incidence of *H. pylori* in cats and their related persons (M: male; F: female).

Sample	Cats						Healthy persons						Diseased persons					
	Domestic			Stray			M			F			M			F		
	No.	P.	%	N0.	P	%	No.	P.	%	No.	P	%	No.	P	%	no.	P	%
Stool	52	16	30.8	18	5	27.8	16	8	50	28	10	35.7	6	4	66	10	8	75
Saliva	45	20	44.4	18	7	38.9	16	6	37.5	28	14	50	6	3	50	10	7	70
Stomach juice	15	2	13.3	10	3	30.3	-	-	-	-	-	-	6	4	66.7	10	8	80
Total	112			46			76			16			30			16		



C:Cats, M: male, F:female

Fig.(2):The incidence of *H. pylori* in cats and their related persons.

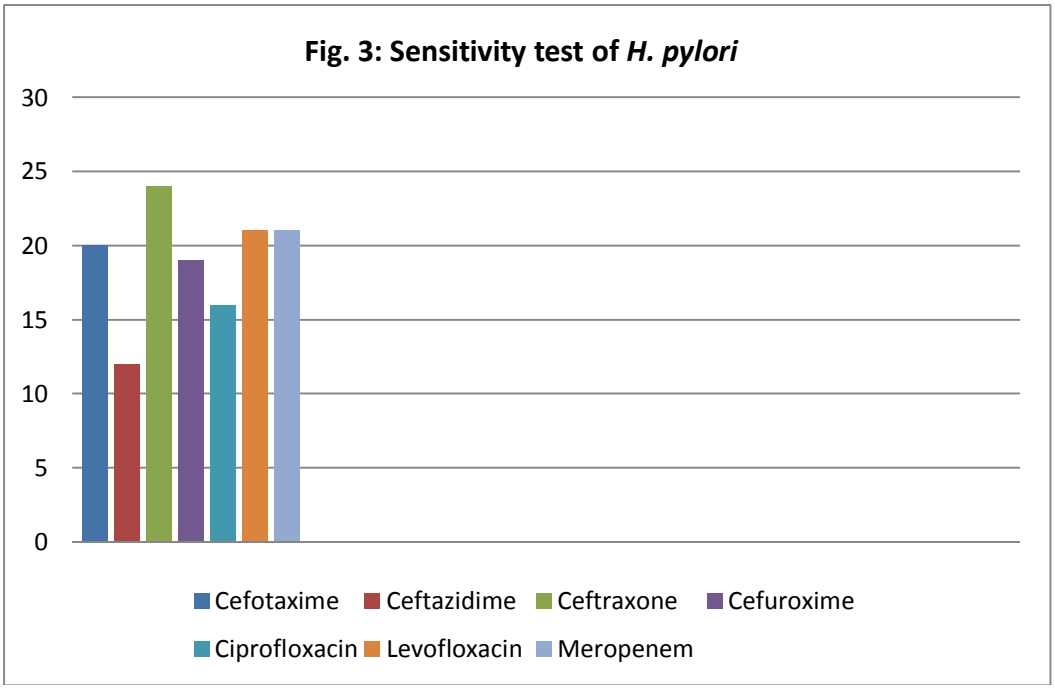


Table 6: 95% Confidence Interval (CI 95%) for the Prevalence rate of *H. pylori* infection in stool, saliva and stomach juice in dogs, cats, and their related persons.

HOST	CI 95% for the Prevalence rate of *H. pylori* infection
Domestic dogs	0.478 ± 0.13448
Stray dogs	0.439 ± 0.23591
Domestic cats	0.333 ± 0.12809
Stray cats	0.275 ± 0.20628
Healthy persons related to dogs	0.191 ±0.1136
Diseased persons related to dogs	0.524 ±0.26161
Healthy persons related to cats	0.432 ± 0.14637
Diseased persons related to cats	0.576 ± 0.2422

4- Discussion.

Incidence of *H. pylori* in Dogs.

From the obtained data in Table 4 revealed that the incidence of *H. pylori* in stool, saliva and stomach juice from domestic dogs ware 45.3%, 45.3% and 53.3%, respectively but 29.4%, 35.3%, and 30.3%, respectively from stray dogs (with an incidence of *H. pylori* in stool, saliva and stomach juice samples of examined dogs of 41.4 %, 42.9% and 50%, respectively).

The incidence of *H. pylori* in all samples of examined dogs was 43.9%. Happonen (1999) found that the prevalence of gastric *Helicobacters* in dogs was between 86 and 100 % in healthy dogs, 61 to 82% in dogs with upper gastrointestinal signs. Wolf (2001) found that the prevalence was higher in dog populations (average of 27.3%). Mercedes *et al.* (2002) concluded that the prevalence of *Helicobacter* spp was observed in 40% of dogs with gastrointestinal signs and in 28% of dogs without gastrointestinal signs. In contrast, Simpson (2000) reported that the prevalence of infection in dogs was: 67% to 86% of clinically healthy dogs and 74% to 80% of dogs presented for investigation of recurrent vomiting. Also, with Neiger and Simpson (2000) reported that the prevalence of infection was dogs is 80.3 %.

Incidence of *H. pylori* in cats.

The data in Table 5 revealed that the incidence of *H. pylori* in stool, saliva and stomach juice from domestic cats ware 30.8%, 44.4% and 22.9%, respectively but 27.8%, 38.9%, and 13.3%, respectively from stray cats (with an incidence of *H. pylori* in stool, saliva and stomach juice samples of examined cats ware 30 %, 42.9% and 20%, respectively). The incidence of *H. pylori* in all samples of examined cats ware 31.7%, this agree with Handt *et al.*(1995) as well as with Dubois (1998) who reported that a natural *H. pylori* infection has been found in the domestic cat.

El-Zaatari *et al.*(1997) failed to isolate *H.pylori* from stray cat**.** Such attitude simulates the results of Yamasaki *et al.*(1998) who reported the isolation of viable *H. pylori* from examined cats reached to 50% of their saliva. Also, Maria *et al.* (2000)found that 34% of gastric juice samples of cats were positive by culture for *H. pylori*. Bob Sherding (2001) who mentioned that prevalence of *H. pylori* were 30% - 100% in stray cats. However, disagree with Geyer *et al.* (1993) who proved that prevalence of gastric *Helicobacter* is more than 90% of healthy pet cats. Handt *et al.* (1994) reported the isolation of viable *H. pylori* from 90% of their gastric juice of domestic cats. Neiger *et al.* (1998) reported that the prevalence of *H. pylori* in cats were between 57% and 100%.Happonen (2003) who reported that the prevalence was ranged between 41% to 100% in healthy cats and 56% to 76% in cats with upper gastrointestinal signs.

The incidence of *H. pylori* in persons related to dogs and cats.

The data in Table 4 revealed that the incidence of *H. pylori* in stool samples from healthy persons related to dogs is 23.9 % (26.3% males and 12.5% females) with CI 95%: 0.239 ± 0.12324 but, in stool samples from persons with gastrointestinal disorders is 64.3 % (60% males and 75% females) with CI 95%: 0.643 ± 0.25098).

In saliva samples from healthy persons, the incidence is 8.7% (7.9% males and 12.5% females) with CI 95%: 0.087±0.08144 buts, in saliva samples from persons with gastrointestinal disorders is 42.9% (50% males and 25% females) with CI 95%: 0.429 ±0.2593. The incidence of *H. pylori* in gastric juice samples from persons with gastrointestinal disorders related to dogs was 62.5% (60% males and 66.7% females). The incidence of *H. pylori* in all samples of all examined persons related to dogs was 30.9 %. Total incidence of examined persons related to dogs was 28.3% males and 33.3% females. These results showed that, females relative infection with *H. pylori* were more than males.

In the present study (table 5) the incidence of *H. pylori* in stool samples from healthy persons related to cats was 40.9 % (50% males and 35.7% females)with CI 95%: 0.409 ± 0.14527. However in stool samples from persons with gastrointestinal disorders is 75 % (66.% males and 75% females) with CI 95%: 0.75 ± 0.21218. In saliva samples from healthy persons related to cats, the incidence is 45.5% (37.5% males and 50% females) with CI 95%:0.455 ± 0.14714 but in saliva samples from persons with gastrointestinal disorders is 62.5% (50% males, 70% females) with CI 95%: 0.625 ± 0.2372.

The incidence of *H. pylori* in gastric juice samples from persons with gastrointestinal disorders related to cats is 75% (66.7% males, 80% females). The incidence of *H. pylori* in all samples of all

examined persons related to cats is 49 %. Total incidence of examined persons related to cats is 41% males and 54.7% females. These results showed that females were more susceptible to *H. pylori* infection than males while persons related to cats probability of *H. pylori* infection more than persons related to dogs.

The presence of bacterial DNA in the oral cavity may be relevant to its transmission(Ierardi *et al.*, 2014). This paper attempts to review the current body of evidence regarding the role of dental plaque, saliva, and periodontal disease in *H. pylori* infection. Anand *et al.* (2014) suggested that the although the way the infection is transmitted is still unclear and interpersonal transmission appears to be the main route. Such attitude simulates the results reported by Blecker *et al.* (1994) who mentioned that the prevalence of *H. pylori* infection inhuman of was 50% –90%. Tsai *et al.* (2005) stated that *H. pylori* has a world-wide distribution as it infects approximately 50% of the world population and its prevalence in healthy asymptomatic persons is 15% - 70%. These results agree with Orderdo *et al.* (1999) who concluded that 40% -50% of adult persons is colonized by *H. pylori* and disagree with Krajden *et al.* (1989) who reported that the prevalence was (40.8%) of the stomach biopsies yielded *H. pylori*.

None of the saliva samples and only one of the dental plaque samples was found positive for *C. pylori*. Nahar et al.(2004) reported that among the culture-positive patients to *H. pylori* (69.5%) were males and (30.4%) were females.

The results in Fig. 3 showed that *in vitro* antibiotic susceptibility of *H. pylori* isolates to different antimicrobial discs are cefotaxime sodium**,** ceftazidime**,** ceftraxone**,** cefuroxime sodium**,** ciprofloxacin**,** levofloxacin**,** meropenem, nitrofurantion and ofloxacin. These results agree with those of other researches (Megraud, 2004; Nicola *et al.*, 2004; Nahar *et al.*, 2004). Boyanova *et al.* (2006) reported that amoxicillin is one of the most commonly used antimicrobial agents for *H. pylori* in recent years, although a high percentage of the isolates were resistant to amoxicillin. Gerrits *et al.* (2002) indicated the resistance to penicillin. McMahon *et al.*(2003) who discussed the relationship among previous antimicrobial use, antimicrobial resistance, and treatment outcomes for *H. pylori* infections. Marco and Antonio (2004) concluded line amoxicillin in his three line of therapy.

Antibiotic sensitivity results show an increased prevalence of *H. pylori* resistant to antibiotics and this agreed with McNulty *et al.* (2002) as antibiotic resistance reduces treatment efficacy, it is time to consider routine susceptibility testing to guide individual patient treatment and surveillance of antibiotic resistance and he noted that there are no published nationally agreed standards for disc diffusion testing of *H. pylori.* Nahar *et al.* (2004) concluded that antibiotic resistance is an emerging problem for the treatment of *H. pylori*-infected patients. Cellini (2014) stated that Bio-film formation in *H. pylori* in the human host also leads to recalcitrance to antibiotic treatment, thus hampering eradication. These lifestyle changes of *H. pylori* allow for a "safe haven" for its survival and persistence according to different ecological niches, and strongly emphasize the need for careful *H. pylori* surveillance to improve management of the infection.

Corresponding author:
Dr. Mohamed Abdel-Raouf Fouda
Gastro-Enterology Center, Mansoura University, Mansoura,Egypt.
E. mail:mohamed.raouf999@yahoo.com

References.

1. Anand, PS., Kamath, KP., Anil., S. (2014):Role of dental plaque, saliva and periodontal disease in *Helicobacter pylori* infection. World J Gastroenterol. 20(19): 5639-53.
2. Blecker, U.,Lanciers, S., Hauser, B., et al.(1994): The prevalence of *H. pylori* positivity in a symptom free population aged 1 to 40 years. J Clin Epidemiol., 47:1095-1098.
3. Bob Sherding, (2001):World Small Animal Veterinary Association Congress (WSAVA) 2001 - Is *Helicobacter* A Problem In Cats.
4. Boyanova, L., Nikolov, R., Lazarova, E., Gergova, G., Katsarov, N., Kamburov, V., Spassova, Z., Derejian, S., Jelev, C., Mitov, I., Krastev, Z. (2006): Antibacterial resistance in *H. pylori* strains isolated from Bulgarian children and adult patients over 9 years. J Med. Microbiol., 55: 65-68.
5. Cellini, L. (2014): *Helicobacter pylori*: a chameleon-like approach to life. World J Gastroenterol. 20(19): 5575- 5582.
6. Christopher, W., Olsen Dec.,(2004): Department of Path biological Sciences, School of Veterinary Medicine, University of Wisconsin-Madison Widespread Zoonotic Potential are Recent Reports of *H. pylori* in domestic cats.
7. De Boer, WA. (1997): Diagnosis of *H. pylori* infection. Review of diagnostic techniques and recommendations for their use in different clinical settings. Scand J Gastroenterol Suppl.; 223:35-42.
8. Rothenbacher, D. and Brenner, H., (2003): Burden of *Helicobacter pylori* and *Helicobacter*-related diseases in developed countries: recent developments and future

implications. Microbes and Infection.; 5:693-703.
9. Dubois, A., Lee, CK., Fiala, N., Kleanthous, H., Mehlman, PT., Monath, T., (1998): Immunization against natural *H. pylori* infection in non-human primates. Infect Immun., 66:4340-4346.
10. Forbes, BA, Sahm, DF, Weissfeld, AS., (2002): Bailey and Scott's Diagnostic Microbiology, 11th ed. Elsevier Science; Chap 39.
11. Fox, JG., (1995): Division of Comparative Medicine, Massachusetts Institute of Technology, Cambridge, USA Non-human reservoirs of *H.pylori*: Aliment Pharmacol Ther.; 9 Suppl 2:93-103.
12. Fox, J. G., M. Batchelder, R. P. Marini, L. Yan, L. K. Handt, X. Li, B. Shames, A. Hayward, J. Campbell, and J. C. Murphy. (1995):*H. pylori*-induced gastritis in the domestic cat. Infect. Immun.; 63:2674-2681.
13. Gerrits, M. M., Schuijffel, D., van Zwet, A. A., Kuipers, E. J., Vandenbroucke-Grauls, C. M. J. E., Kusters, J. G. (2002): Alterations in penicillin-binding protein 1A Confer Resistance to {beta}- Lactam Antibiotics in *Helicobacter pylori*. antimicrob. Agents Chemother., 46: 2229-2233.
14. Geyer, C., Colbatzky, F., Lechner, J., Hermanns, W.(1993): Occurrence of spiral-shaped bacteria in gastric biopsies of dogs and cats. Vet. Rec., 133: 18-19.
15. Goodwin,C. S., McCulloch, R. K., Armstrong, J. A., *et al.* (1985): Unusual cellular fatty acids and distinctive ultra structural in anew spiral bacterium (*C.pyloridis*) from the human gastric mucosa. J. Microbiol.; 19:257-267.
16. Hachem, C. Y., Clarridge, J. E., Evans, D. G., Graham, D. Y.(1995): Comparison of agar based media for primary isolation of *H. pylori*. J. Clin. Pathol., 48:714-716.
17. Handt, L. K., Fox, J. G., Dewhirst, F. E., Fraser, G. J., Paster, B. J., Yan, L. L., Rozmiarek, H., Rufo, R., Stalis, I.(1994): *H. pylori* isolated from the domestic cat: public health implications. Infect. Immun. 62(6):2367-2374.
18. Handt, L. K., Fox, J. H., Stalis, R., Rufo, G., Lee, J., Linn, X. Li, H., Ierardi, E., Goni, E., Losurdo, G., Di Mario, F. (2014): *Helicobacter pylori* and nonmalignant diseases. *Helicobacter* spp. Suppl 1: 27-31.
19. Happonen, (2003): Canine and Feline Gastric *Helicobacters*: Diagnosis and Significance in Chronic Gastritis 28th Congress of the World Small Animal Veterinary Association (WSAVA) in Bangkok, Thailand, October 24-27.
20. Hazell, SL., Borody,TJ.,Ga,l A., *et al.* (1987): *C. pyloridis* gastritis, detection of urease as a marker of bacterial colonization and gastritis. Am J Gastroenterol., 82:292-296.
21. Jerris, R. C., Murrary, P. R., Boran, E. J., Pfaller, M. A., Tenover, F. C., Yolken, R. H. (1995): *Helicobacte*r. In: Manual of Clinical Microbiology. American Society of Microbiology. Washington, D.C.; 6th Edition. pp. 492-498.
22. Krajden, S., Fuksa, M., Anderson, J., Kempston, J., Boccia, A., Petrea, C., Babida, C., Karmali, M., Penner, JL. (1998):Examination of human stomach biopsies, saliva, and dental plaque for *C. pylori*. J Clin Microbiol., 27(6):1397-1398.
23. Kleanthous (1995): Characterization of feline *H. pylori* strains and associated gastritis in a colony of domestic cats. J. Clin. Microbiol., 33:2280-2289.
24. Klein, PD., Malaty, HM., Martin, RF., *et al.* (1996): Noninvasive detection of *H. pylori* infection in clinical practice: the 13C urea breath test. Am J Gastroenterol., 91: 690-694.
25. Lavelle, J.P., Landas, S., Mitros, F. A., Conklin, J. L., (1994): Acute gastritis associated with spiral organisms from cats. Dig. Dis. Sci., 39: 744-750.
26. Lopez -Brea, M., Alarcon, T., Megraud, F. (1997): Diagnosis of *H. pylori* infection. Current opinion in Gastroenterology Suppl., 13:13—19.
27. Malfertheiner, P., Pieramico, O. (1992): *H. pylori*: Seven Gustovsson. The Stomach, 12:297-311.
28. Marco, R.M.D., Antonio, C. M.D. (2004): Eradication of *Helicobacter pylori*: A clinical uptake. Meds cape General Medicine., 6(1):e 27.
29. Maria. I., Esteves, M. D., Schrenzel, R. P., Marini, N. S., Taylor, S. Xu., Susan, H., Yan, F., Zeli, S., James G. Fox, (2000): *H. pylori* gastritis in cats with long-term natural infection as a model of human disease Am. J. of Pathology., 156:709-721.
30. McMahon, B. J., Hennessy, T. W., Bensler, J. M., Bruden, D. L., Parkinson, A. J., Morris, J. M., Reasonover, A. L., Hurlburt, D. A., Bruce, M. G., Sacco, F., Butler, J. C.,(2003): The relationship among previous antimicrobial use, Antimicrobial Resistance, and Treatment Outcomes for *H.pylori* Infections. Ann Intern Med 139: 463-469.
31. McNulty, C., Owen, R., Tompkins, D., Hawtin, p., McColl, K., Price, A., Smith, G., Teare, L. H. (2002): *H. pylori* susceptibility testing by disc diffusion. J. Antimicrob. chemo., 49,601-609.

32. Megraud, F.(2004): *H. pylori* antibiotic resistance: prevalence, importance, and advances in testing. Gut., 53: 1374-1384.
33. Mercedes, G., Sainz, A., Delgado, J., Rodríguez-Franco, F. (2002): Hospital Clínico Veterinario, Facultad de Veterinaria de la Universidad Complutense de Madrid Prevalence of *Helicobacter* spp in Canine Gastric Biopsies Using Urease Test and Relationship with Gastrointestinal Signs World Small Animal Veterinary Association Congress (WSAVA) 2002.
34. Mobley, H.L. T. (1997): *H. pylori* factors associated with disease development. Gastroenterology 113:21-28.
35. Nahar, S., Mukhopadhyay, A. K., Khan, R., Ahmad, M. M., Datta, S., Chattopadhyay, S., Dhar, S. C., Sarker, L., Engstrand, S. A., Berg, D. E., Nair, G. B., Rahman, M. (2004):Antimicrobial susceptibility of *H. pylori* strains isolated in bangladesh.J. Clin. Microbiol., 42(10): 4856 - 4858.
36. Neiger, R., Dieterich, C., Burnens, A., Waldvogel, A., Corthesy-Theulaz, I., Halter, F., Lauterburg, B., Schassmann, A. (1998): Detection and prevalence of *Helicobacter* infection in pet cats. J Clin. Microbiol., Vol. 36, 3:634-637.
37. Nicola, C. E., Roberts, J. O., Jacqueline, X., Mark, W., Kenneth, D. (2004): *H. pylori* antibiotic resistance patterns and genotypes in adult dyspeptic patients from a regional population in North Wales. J. Antimicrobial. Chemotherapy, 54, pp: 435-440.
38. Orderdo, G., Vaira, D., Halton, J. (1999): *H. pylori* in children with peptic ulcer and their families. Dig. Dis. Sci., 36: 572-579.
39. Radin, M.J., Eaton, K.A., Krakowka, S., Morgan, D.R., Lee, A., Otto, G., Fox, J. (1990): *H.pylori* gastric infection in gnotobiotic beagle dogs. Infect. Immun., 58: 2606-2612.
40. Simpson, K. (2000):Gastric *Helicobacter* species infection in dogs and cats. Practive, 21(8):427-435.
41. Suerbaum, S., Michetti, P.(2002):*Helicobacter pylori* infection. N Engl J Med.; 347: 1175–1186.
42. Taylor, D. N., Blaser, M. J. (1991): The epidemiology of *H. pylori* infection. Epidemiol. Rev., 13: 42-59.
43. Topley, Wilson (1998): Microbiology and Microbiological Infections, Ninth edition Vol.3.
44. Toracchio, S., Marzio, L. (2003): Primary and secondary antibiotic resistance of *H. pylori* strains isolated in central Italy during the years 1998-2002. Dig. Liver Dis. 35:541-545.
45. Tsai, C. J., Perry, S., Sanchez, L., *et al.* (2005): *H. pylori* infection in different generations of Hispanics in the San Francisco Bay Area. Am.J. Epidemiol., 162(4): 351-357.
46. Wolf, A. (2001): Emerging Diseases Human Concerns Congress of the World Small Animal Veterinary Association WSAVA.
47. Yamasaki, K., Suematsu, H., Takahasi, T. (1998): Comparison of gastric lesions in dogs and cats with and without gastric spiral organisms. J. Am. Vet.Med. Assoc., 212: 529-533.

11/16/2014