

# Prevalence of sexually transmitted infections and enteric protozoa among homosexual men in western Sicily (south Italy)

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## Key words

Homosexual men • Sexually transmitted infections • Enteric protozoa • Western Sicily

## Summary

**Introduction.** In recent years an increase in the number of sexually transmitted infections (STIs) in men who have sex with men (MSM) has been reported in different industrialised countries. Because few epidemiological data on the STIs/MSM population in Sicily are available, a survey was conducted to assess the prevalence of STIs/enteric protozoa and risky sexual behaviours among MSM in western Sicily.

**Methods.** In 2010, 74 MSM with median age of 30 years old, were recruited via networks. All participants to the study were interviewed by anonymous self-administered questionnaire in order to collect social/demographic information, clinic data and STI-related risky sexual behaviours. After completing the questionnaire, blood samples were collected to determine HIV, HCV, HHV8 and *Treponema pallidum* antibodies; presence of *Giardia duodenalis* and *Cryptosporidium parvum* was also investigated in faecal samples by immunofluorescence assay.

**Results.** HIV, HHV8, *T. pallidum* and *Giardia* prevalence were 8.1%, 16.2%, 21.6% and 16.4% respectively; all patients were negative for HCV and *Cryptosporidium* infections. The median values of sexual anal intercourse and oral sex per week were 2 and 1, respectively. 7% of participants always had unprotected anal sex, 50.7% sometimes used condom during sexual anal intercourse and 42.3% always had protected anal sex. All MSM-HIV+ and 7 (43.7%) syphilis seropositives were unaware of their own infection.

**Discussion.** MSM in western Sicily are a high risk group for important STIs. It seems necessary that continuous interventions for preventing HIV/AIDS and other STIs and for improving the level of knowledge of symptoms are needed.

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## Introduction

Sexually transmitted infections (STIs) are a major public health problem in Europe. In particular, sexual activity has been shown to be the primary mode of transmission for several important viral, bacterial and parasitic infections among men who have sex with men (MSM) throughout the world [1-3].

Between the 1980s up to the 1990s, a reduction in the incidence of STIs was observed among MSM likely due to behavioural change occurring in response to the emergence of HIV/AIDS [4]. However, this attention has not been maintained after the diffusion of the Highly Active Antiretroviral Therapy (HAART) and the proportion of homosexual men reporting "unsafe sex", often measured as unprotected anal intercourse (UAI) with casual partners, has increased since the mid-1990s [4].

As consequence, many States are now observing increases in rates of several STIs and outbreaks of syphilis and lymphogranuloma venereum infection were reported among the MSM population living in different industrialized Countries [5, 6].

The World Health Organization (WHO) estimates 12 million new syphilis cases worldwide annually, of which 140,000 in western Europe where the infection has re-emerged both in homosexual men and in het-

erosexuals [2, 7, 8]. An increase of cases among MSM has been also found in Italy, where syphilis cases have increased tenfold from 2000 to 2007 [9], Norway [10], Scotland [11] and in Germany [12].

Moreover, recent studies have shown that in Canada [13], the United States [14], Australia [15] and China [2] MSM represent the risk group diagnosed most frequently with HIV infection. In the United States, the MSM population accounted for the majority of new HIV infections in 2006 [16] so president Obama's released National HIV/AIDS Strategy calls for more emphasis on addressing the HIV prevention and care needs of MSM [17].

Several other infectious agents, such as *Giardia duodenalis*, *Cryptosporidium parvum*, Hepatitis C virus (HCV) and Human Herpesvirus 8 (HHV8) have been found to cause diseases among MSM, probably as consequence of both unprotected oral sex and other sexual practices (unprotected anal sex, rimming, fisting) that may facilitate sexual transmission because of bleeding (visible or not) during sex [18].

According to these considerations, high rates of enteric protozoan parasitism, related to sexual activity, have been reported among MSM in metropolitan areas worldwide [3] whereas, in western Sicily, Perna et al. [19] have found HHV8 seroprevalence rates among HIV positive and negative gay men of 62% and 22%, respectively.

Considering that a general lack of knowledge exists on MSM-STIs in Sicily, the aim of the present study was to assess STI seroprevalence, risk behaviours and economic-social factors possibly involved in the transmission of STIs among MSM population living in western Sicily.

## Methods

### STUDY POPULATION

Seventy four MSM were recruited mainly via internet and gay-pub between February 2010 and December 2010. Eligibility criteria for participation were as follows: being aged 18 years or older and living in western Sicily for at least 6 months.

All MSM were informed by a letter on the nature and purpose of the study and, after obtaining a written informed consent, a self-administered questionnaire was submitted to each individual. Participation in this study was completely voluntary and anonymity was granted using the initials of the first and last name followed by the year of birth or by pseudonyms.

### QUESTIONNAIRE AND BIOLOGICAL SAMPLES COLLECTION

An anonymous self-administered questionnaire was used to collect the following data: socio-demographic and clinic information (age, civil status, highest education level, HIV status, diarrhoeic symptoms in the last month, travel in developing countries, care of hygiene hands, diagnosis of STIs, previous blood transfusion); drug-using (injecting use) and sexual behaviours (age of homosexual experience, sexual orientation, number of sexual partners in the last year, consistent condom use during anal intercourse, stable or occasional partner in the past month). Sex was defined as oral and anal and each patient also was asked about their attendance for week.

By means questionnaire we also collected information on economic (no-temporary job and temporary-job, educational level) background.

No incentives were provided for completing the questionnaire.

After completing the questionnaire, blood and stool samples were collected.

No participants refused serologic testing; 8 MSM not provided stool samples for embarrassment. All study participants had the opportunity to receive their test results with post-test counselling within one week after the blood draw and providing faecal samples.

### SERUM SPECIMENS

All participants (n = 74) were processed for HIV, HCV, HHV8 and *Treponema pallidum* infections. Serum samples were first tested for anti-HIV1+2 and anti-HCV by chemiluminescence immunoassay (VITROS, Ortho-Clinical Diagnostics, US) and positive results were confirmed by Chiron Riba HIV1/HIV2, (Ortho-Clinical Diagnostics) and Chiron Riba HCV 3.0 SIA (Ortho-Clinical Diagnostics). At same time all patients were analyzed for HHV8 antibodies by using an immunofluorescence

assay (IFA) based on BCBL-1 cell line as previously described [19]; samples were considered positive if reactive at dilution  $\geq 1:120$ . All serum specimens were tested for total anti-*T. pallidum* antibodies using a sandwich enzyme-linked immunoassay (ELISA) (Radim, Pomezia, Italy). Specimens with positive ELISA reactions were tested for non-treponemal rapid plasma reagin (RPR test) antibodies (Pulse Scientific, Burlington, Ontario) and those with RPR titre  $< 1:8$  underwent confirmatory testing in the *T. pallidum* haemagglutination assay (TPHA) (ASI TPHA test kit, Arlington Scientific, USA). Serum samples with a positive ELISA and a RPR titre  $\geq 1:8$  were considered indicative of recent syphilis infection while those with a positive ELISA, a RPR titre  $< 1:8$ , and a positive TPHA reflected past infection.

### STOOL SAMPLES

Eight patients of the 74 men invited to participate in the study not provided the stool sample. The faecal samples (n = 66) were examined by formol-ether concentration. Briefly, 2-3 g of faeces were suspended in 10mL of saline solution and filtered through a surgical gauze. The filtered was then centrifuged at 1500 rpm for 2 min. The supernatant was decanted and the pellet was suspended in 7 ml of formalin at 7% and 3 ml of ethyl acetate. The sample was mixed for 3 min and centrifuged at 1500 rpm for 2 min. The supernatant was discharged and the final pellet was processed by immunofluorescence assay (Merifluor *Cryptosporidium/Giardia* assay; Meridian Biosciences) for the simultaneous detection of *C. parvum* and *Giardia duodenalis*. The slides were observed with an epifluorescence microscope at 400 x magnification for the detection of FITC-mAb labeled oocysts/cysts. Presence of stained oocysts/cysts was identified according to morphology.

### STATISTICAL ANALYSIS

The questionnaire responses were entered in a electronic worksheet (Microsoft Excel). Absolute and relative frequencies were calculated for qualitative variables, while quantitative variables were summarized as median (range). Odds ratio (OR) with 95% confidence intervals (95% CIs) were calculated by univariate logistic regression analyses.

The significance level chosen for all analysis was 0.05, two-tailed. All the data were analyzed using the R statistical software package [20].

## Results

### SOCIO-DEMOGRAPHIC DATA

The median age of the 74 screened individuals was 30 years (range 18-56 years). Most participants (n = 72) were Italians; the 2 MSM non-Italians (both living in Palermo city) were a French man and an Argentinian, respectively. Regarding education level, 4 (5.5%) had attended a low secondary school, 31 (42.5%) had graduated from high secondary school, 34 (46.6%) had attended University degree, and 4 (5.5%) reported University

Specialty. Of the processed MSM, 70 (94.6%) were single, 1 (1.4%) was divorced and 3 (4.1%) were cohabiting partners. Twenty seven (36.5%) respondents declared no-temporary job, 33 (44.6%) temporary-job, 11 (14.9%) were no employed and 2 (2.7%) retired whereas 1 (1.3%) subject did not answer to this question.

### SEXUAL BEHAVIOUR

As shown in Table I, among 74 MSM who participated in the study 11 (15.1%) were bisexual men. The median age of homosexual experience was 9 years (range:1-32). A small proportion of the study population (7%) never used condom during sexual anal intercourse and slightly less than half (50.7%) sometimes used it while 30 participants (42.3%) always used it.

The median value of total number of male sex partners in the last year was 5 (range:1-100). The median value of anal sexual intercourse and oral sex per week were 2 (range: 0-5) and 1 (range: 0-6) respectively. Most of the participants (60.6%) declared for engaging indifferently in insertive and receptive anal intercourse while 16 (22.5%) and 12 (16.9%) had only insertive or receptive anal sex, respectively. Forty one MSM (55.4%) stated they had used heavy drugs during the previous year.

No independent variable was statistically significantly associated with giardiasis or being seropositive for HIV or syphilis infection (Tab. II).

### SEROPREVALENCE OF HIV, HHV8, HCV, T. PALLIDUM

Among the participants, 6 MSM (8.1%) had been diagnosed with HIV-1 infection, 16 (21.6%) with syphilis and 12 (16.2%) with HHV8 infection; all patients were negative for HCV. None patient had a syphilis/HIV coinfection.

Seven (43.7%) syphilis seropositives and all HIV-1 positive (100%) were unaware of their own infection.

### PREVALENCE OF GIARDIA/CRYPTOSPORIDIUM

Of the 66 patients who submitted faecal samples, a total of 11 (16.6%) was positive for *G. duodenalis*. All MSM with giardiasis were treated orally with anti-giardial therapy; the treatment reached 100% of efficacy because no cysts were found in the faeces after cure. No *Cryptosporidium parvum* was detected in any of the processed samples, including the HIV- positive group of MSM.

### Discussion

In the last decades, industrialized Countries have considered MSM as a high risk group for infections related with sexual activities. However, it seems that a such attention is gradually declining as consequence of the HAART treatment.

The present study shows that MSM living in developed Countries may continue to be a very high risk group, since up to one third of them has had diagnosed one between syphilis and HIV and only half of these subjects were aware of their own seropositivity.

Both the HIV prevalence and unawareness of the infection are consistent with data reported in literature and a recent survey carried out in several large U.S. cities has indicated approximately one in four MSM surveyed in social venues is infected with HIV, and nearly 50% of them are unaware of their HIV seropositivity [21]. The high proportion of MSM unaware of their seroconversion must be considered a serious public health concern, since these MSM can account for a large majority of estimated new HIV transmissions, as reported in the United States, and persons unaware of their HIV infection often leave out substantial steps to reduce their risk behaviours, increasing the risk of HIV transmission [22].

Tab. I. Sexual behaviours/characteristics of the 74 MSM.

Bisexual orientation, n (%)		
Yes	11	(15.1)
No	62	(84.9)
Years since the first homosexual experience, median (range)		
	9	(1-32)
Condom use, n (%)		
Never	5	(7)
Sometimes/often	36	(50.7)
Always	30	(42.3)
Total number of male sex partners in the last year, median (range)		
	5	(1-100)
Sexual intercourse per week, median (range)		
	2	(0-5)
Oral intercourse per week, median (range)		
	1	(0-6)
Anal intercourse, n (%)		
Insertive	16	(22.5)
Receptive	12	(16.9)
Both insertive and receptive	43	(60.6)
Heavy drugs consumption in the last year, n (%)		
Yes	41	(55.4)
No	33	(44.6)

\* Percentages may not total 100% due to missing data

Tab. II. Risk factors associated with HIV, syphilis and giardiasis.

	HIV or syphilis seropositivity OR (95% CI)	p-value	Giardiasis seropositivity OR (95% CI)	p-value
Age, in years	1.01 (0.95-1.06)	0.82	0.98 (0.91-1.06)	0.72
Residence (Palermo vs Other)	0.51 (0.16-1.59)	0.25	0.59 (0.14-2.62)	0.33
Highest study level (University or Specialty vs secondary or tertiary school)	0.68 (0.25-1.87)	0.46	0.44 (0.12-1.69)	0.23
Years since the first homosexual experience	1.04 (0.96-1.19)	0.39	0.98 (0.88-1.08)	0.64
Bisexual orientation (yes vs no)	0.47 (0.09-2.36)	0.36	0.19 (0.01-3.46)	0.75
Anal intercourse, n (%)				
Both insertive and receptive	1		1	
Insertive	0.86 (0.23-3.2)	0.82	0.53 (0.10-2.8)	0.45
Receptive	2.58 (0.69-9.61)	0.16	-	-
Total number of male sex partners in the last year	0.99 (0.96-1.02)	0.58	1 (0.97-1.04)	0.99
Total number of sexual intercourse per week	1.12 (0.78-1.62)	0.53	1.03 (0.65-1.62)	0.90
Total number of oral intercourse per week	1 (0.71-1.42)	0.96	1.1 (0.73-1.67)	0.65
Condom use				
Always	1		1	
Sometimes/often	0.58 (0.2-1.66)	0.31	0.99 (0.24-4.12)	0.99
Never	1.15 (0.17-7.99)	0.89	3.83 (0.48-30.7)	0.21
Heavy drugs consumption in the last year (yes vs no)	0.73 (0.26-1.99)	0.54	0.52 (0.13-1.95)	0.32
Drinking tap water				
Never	1		1	
Yes, sometimes	0.45 (0.12-1.6)	0.21	3.7 (0.78-17.38)	0.10
Yes, always	-	-	11 (0.54-223.9)	0.12
Eating raw vegetables				
Never	1		1	
Yes, sometimes	0.87 (0.27-2.81)	0.81	5 (0.57-44.6)	0.15
Yes, always	0.33 (0.03-3.33)	0.35	3 (0.16-55.66)	0.46
Hand washing (times per day)	1.08 (0.93-1.25)	0.30	1.03 (0.87-1.23)	0.73

The high prevalence of syphilis among MSM participants to this study confirms observations from previous studies [1, 9-12] indicating that a predominant mode for its transmission is through homosexual intercourse. From the epidemiologic point of view, the increase of the disease is an alarming datum because there is strong evidence that syphilis facilitates HIV transmission [23] and the infection is frequently asymptomatic. In our study, the finding that none patient have a simultaneous HIV/syphilis infection may be related to the high probability that they were aware of their health status and, consequently, did not accept our invitation. HHV8 seroprevalence rates among MSM, even if it is lower as regards the results of a previous study [19] carried out in western Sicily, confirm that HHV8 infection is widespread in our geographical area particularly in individuals at risk for STIs. Some authors have found that this high prevalence of HHV8 among homosexual men could be due to a transmission of the virus via saliva during oral-genital sex [24].

Differently from HIV, syphilis and HHV8 seroprevalences, our results show that no MSM was HCV seropositive. This datum suggests that the risk of HCV acquisition through sexual routes is very low. According to this finding, in England, Scott et al [25] screened for HCV 2,309 MSM and found an anti-HCV prevalence lower than 1% both in the HIV-positive and HIV-negative groups.

Intriguingly, although all tested MSM were asymptomatic for intestinal diseases, the results of our study (prevalence of giardiasis of 16.6%) support the experience of others authors regarding high prevalence of intestinal parasites infection as giardiasis in the homosexual population [26, 27]. The high rate of *G. duodenalis* infection has been associated with oral, anal, faecal-oral contact (i.e. rimming) and oral-genital sexual practices common among MSM. Considering the increasing importance of those parasites in acquired immunodeficiency syndrome patients we think that MSM-HIV positives always should be submitted for parasitologic examinations, particularly with regard to enteropathogens like *G. duodenalis*.

Otherwise, the total absence of *Cryptosporidium parvum* could be justified both by the low presence of this parasite in western Sicily [28] and by the small sample of participants to the study.

Finally, this study may have some limitations. The small sample of MSM who agreed to participate in this study and the use of self-reported data represent the first two limitations. However, both of these are also important results that highlights the strong cultural resistance and the embarrassment to homosexuality in the examined Sicilian context. Accordingly, the absence of associations between STIs and risk factors, that are frequently reported by the international litera-

ture, can be explained by considering that some high risk behaviours may have been underreported, leading to potential underestimation of any associations. As third point, a self-selection bias may have restricted the generalizability of our findings.

## Conclusion

Despite some possible limitations, the data of prevalence obtained in the present study provide useful information for health promoters because confirm that MSM living

in developed Countries can be a very high risk group for important viral, bacterial and parasitic infections. The major challenge for the future is to reduce the rates of new infections in developed geographic areas where homosexuality is still lived with shame. Behavioural prevention remains central to halt the spread of these STIs among gay and bisexual men, so health promoters need to collaborate with those men to find effective ways to minimise risk of infection. Providing culturally and contextually appropriate messages is essential to help persons at risk avoid contracting HIV/STIs and to help who are infected to avoid transmitting the infection.

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