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Genital *Chlamydia trachomatis* infections among adolescents in a high-incidence area in Norway: genotypes, prevalence, early sexual behaviour and testing patterns – a cross-sectional study

The Finnmark High School Study (FHSS)

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A dissertation for the degree of

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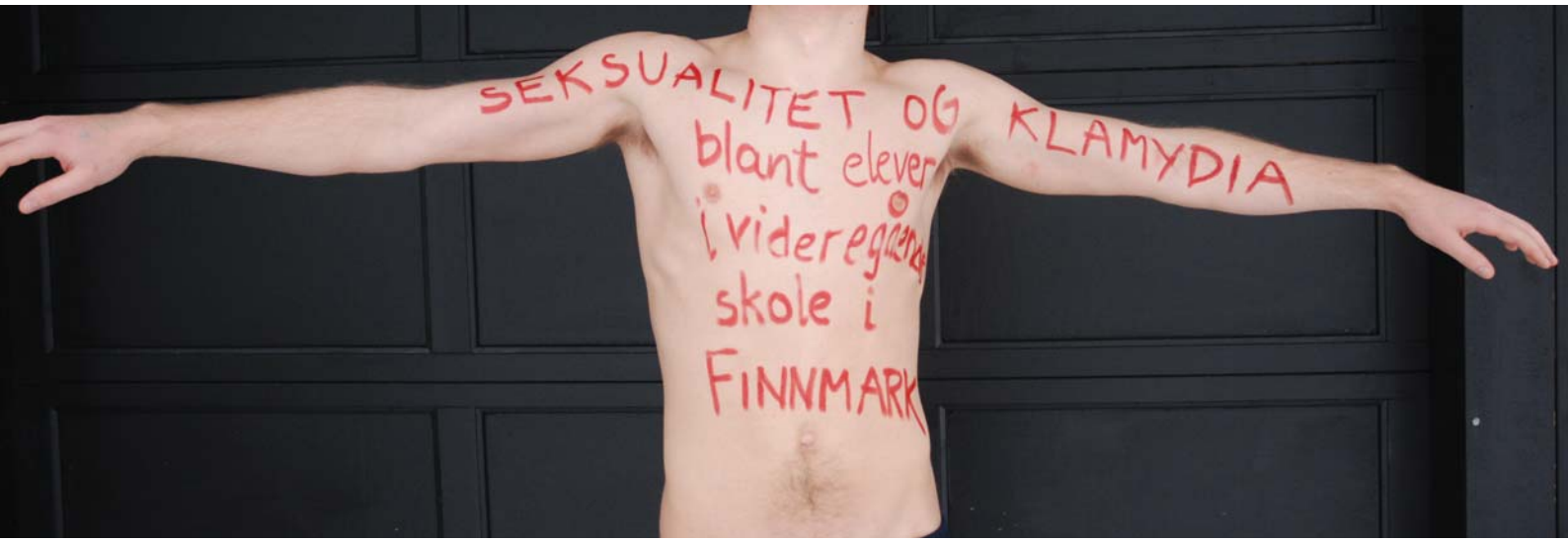

'Sexual behaviour and chlamydia among high school students in Finnmark'

Comments heard during data collection

'Seriously, do you really think I'm just going to sit here and do boring school work while the others are answering that extremely interesting sex-quest – hello, I've changed my mind, email me that quest right away!'

'This questionnaire is so useful to sum up my life experiences'.

'What the f… has education and religion got to do with having a chlamydia infection?'

'What is the problem with having something that doesn't ever show itself?'

'How come you ask me - a boy of only 17 - if I've ever been with a prostitute?'

'Why do you only test our urine samples for chlamydia? You should check for everything!'

Preface

During my years as senior physician at the Regional Centre for Infection Control at the University Hospital of North Norway, I was often approached by colleagues and by representatives from the national health authorities at meetings and conferences who inquired about the 'chlamydia epidemic' in Finnmark county as indicated by surveillance data. The questions would commonly be accompanied by humorous suggestions of reasons for the high chlamydia rates. Every spring, the Norwegian Institute of Public Health would publish their annual chlamydia report that listed priority tasks in the field of chlamydia prevention. 'Increased knowledge about the chlamydia epidemiology in Finnmark' was usually included on that list, but no relevant research studies were initiated. Eventually, I was ready to do my PhD. I realised that this was my opportunity to study genital chlamydia infections among young people in Finnmark and I started planning my PhD project. After two years of applying for funding and permissions, we finally set off to Finnmark in September 2009. We carried boxes and suitcases filled with urine sample transport tubes, disposable gloves and laboratory forms and were enthusiastically received by students and staff in 5 high schools. It turned out to be a fantastic journey. This thesis includes three papers from the Finnmark High School Study.

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Study participants in 5 high schools in Finnmark as there would be no study without you!

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Tromsø, August 23rd, 2013

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Summary

Chlamydia trachomatis is the most commonly reported curable sexually transmitted infection in Western high-income countries and can cause severe female reproductive tract morbidity. Despite extensive control efforts, chlamydia rates have increased in most countries since the mid-1990s. Young persons and especially adolescent girls have the highest infection rates. In general, girls are tested far more frequently than boys. High-resolution genotyping provides detailed information on the molecular epidemiology and genetic diversity of *C. trachomatis*.

In this thesis, we investigated; i) *C. trachomatis* genotype distribution and genetic diversity using MLST (multilocus sequence typing) and *ompA* genotyping in Finnmark, a high-incidence area in Norway, ii) associations between early sexual behaviour and prevalent chlamydia infection, and iii) demographic and sexual behaviour factors associated with chlamydia testing in a high school based screening and previous clinic based testing, among girls and boys aged 15-20 years who participated in the Finnmark High School Study conducted from September to November 2009.

We detected a large genetic diversity, multiple novel sequence types and alleles by MLST, and an atypical genovar distribution with predominance of G in a previously unmapped area. *C. trachomatis* genetic diversity in rural Finnmark and two other urban areas was similar. Chlamydia prevalence in sexually active girls was 7.3% and in boys 3.9%. Girls had earlier sexual debut and were more sexually active at a younger age and thus had a different risk profile from boys which may contribute to higher prevalence. Threefold more girls than boys reported previous clinic based testing which was associated with known chlamydia risk factors. School based screening reached 93% of participants and was associated with factors unknown to increase risk thus suggesting other motives. Half of infections were detected in those only tested at school. We confirmed the efficiency of school based screening to increase testing and detect hidden infections and we thus suggest this approach to be tried as a complement to other chlamydia control strategies in selected high-morbidity areas in Norway.

Sammendrag

Chlamydia trachomatis er den hyppigst rapporterte seksuelt overførbare infeksjonen i vestlige land. Ubehandlet infeksjon hos kvinner kan føre til svangerskap utenfor livmoren og barnløshet. Tross omfattende kontrolltiltak har forekomsten av klamydiainfeksjoner økt i de fleste land siden midten av 90-tallet. Unge personer og særlig kvinner har høyest forekomst. Jenter tester seg generelt mer enn gutter. Genotypimetoder med høy oppløselighet kan gi kunnskap om molekylærepidemiologien og den genetisk diversiteten av *C. trachomatis.*

I denne avhandlingen har vi undersøkt; i) distribusjon og genetisk diversitet av *C. trachomatis* genotyper ved bruk av MLST (multilokus sekvenstyping) og *ompA*-typing i Finnmark som har den høyeste insidensraten av klamydia i Norge, ii) om kjønnsforskjeller i tidlig seksualatferd er relatert til prevalent klamydiainfeksjon, og iii) om demografiske faktorer og seksualatferd har betydning for deltakelse i en klamydiascreening i videregående skole og for tidligere testing i klinisk praksis blant jenter og gutter i alderen 15-20 år som deltok i en forskningsstudie ved fem skoler i Finnmark fra september til november 2009.

Vi påviste stor genetisk diversitet, mange nye alleler og sekvenstyper ved MLST, samt en atypisk genovarfordeling med predominans av G i et ikke kartlagt område. Genetisk diversitet var lik i Finnmark og to større byer. Klamydiaprevalens hos seksuelt aktive jenter var 7,3% og hos gutter 3,9%. Jenter hadde lavere seksuell debutalder og var tidligere mer seksuelt aktive enn gutter. Ulik risikoprofil kan bidra til å forklare kjønnsforskjeller i prevalens. Tre ganger flere jenter enn gutter rapporterte tidligere testing i klinisk praksis, mens testraten var 93% for begge kjønn i skolescreeningen. Tidligere testing var assosiert med kjente risikofaktorer for klamydia, mens deltakelse i screeningen var assosiert med faktorer som vanligvis ikke er knyttet til infeksjonsrisiko. Dette tyder på at andre motiver var viktige for deltakelse i skole-screeningen. Halvparten av infeksjonene ble påvist blant personer som kun testet seg på skolen. Vi bekreftet at skolescreening øker testing og påviser et større infeksjonsreservoar. Vi foreslår derfor at skolescreening utprøves i selekterte områder med høy klamydiaforekomst som et tillegg til andre forebyggende tiltak mot klamydiainfeksjoner i Norge.

List of papers

This thesis is based on the three following papers, which will be referred to in the text by their Roman numerals:

I. **Gravningen K**, Christerson L, Furberg AS, Simonsen GS, Ödman A, Herrmann B. Multilocus sequence typing of genital *Chlamydia trachomatis* in Norway reveals multiple new sequence types and a large genetic diversity. Plos One 2012; 7:e34452. doi:10.1371/journal.pone.0034452 PONE-D-11-25772 [pii].

II. **Gravningen K**, Furberg AS, Simonsen GS, Wilsgaard T. Early sexual behaviour and *Chlamydia trachomatis* infection - a cross-sectional study on gender differences among adolescents in Norway. *BMC Infectious Diseases* 2012;**12**:319. doi:10/1186/1471-2334-12-319.

III. **Gravningen K**, Simonsen GS, Furberg AS, Wilsgaard T. Factors associated with *Chlamydia trachomatis* testing in a high school based screening and previously in clinical practice: a cross-sectional study in Norway. *BMC Infectious Diseases* 2013, **13**:361. doi:10.1186/1471-2334-13-361

Abbreviations

CASI	Computer-assisted self-interview
CI	Confidence interval
CDC	Centers for Disease Control and Prevention (Georgia, US)
C. trachomatis	*Chlamydia trachomatis*
DNA	Deoxyribonucleic acid
ECDC	European Centre for Disease Prevention and Control (Sweden)
FHSS	Finnmark High School Study
FVU	First-void urine
IR	Incidence rate
MLST	Multilocus sequence typing
MOMP	Major outer membrane protein
NAAT	Nucleic acid amplification test
NIPH	Norwegian Institute of Public Health
nvCT	New Swedish mutated variant of *C. trachomatis*
ompA	Gene coding for major outer membrane protein (MOMP)
OR	Odds ratio
PCR	Polymerase chain reaction
SNP	Single nucleotide polymorphism
ST	Sequence type of *C. trachomatis* based on MLST
STI	Sexually transmitted infection
UNN	University Hospital of North Norway
WGS	Whole-genome sequencing

1. Introduction

1.1 Bacteriology

Chlamydia trachomatis is a small (1,000 kB) obligate intracellular bacterial pathogen with a specialised biphasic developmental cycle. The bacterium effectively conceals its antigenic profile from the immunity system by replicating in an intracellular vacuole and then moving between two hosts in the non-replicative form. It belongs to the order *Chlamydiales,* the family *Chlamydiaceae,* and the genus *Chlamydia,* which includes *C. trachomatis* that has humans as its only reservoir. *C. trachomatis* comprises two biovars: the trachoma biovar that includes ocular and urogenital strains causing localised infections of the epithelial surface of conjunctiva or genital mucosa, and the lymfogranuloma venereum biovar that can spread systemically through the lymphatic system causing genital ulcer disease. Most *C. trachomatis* strains possess a cryptic plasmid of 7.5 kB that mostly shares the same evolutionary history as their chromosomes and is putatively linked to virulence [1].

1.2 Clinical course

C. trachomatis has a long infectious period with less than half of untreated infections resolving spontaneously within a year [2, 3]. Repeat infections in adolescents are common suggesting limited development of immunity following a first infection [4-7]. More than 95% of chlamydia infected women and men in population based studies report no symptoms [8]. In women, major clinical manifestations include urethritis and cervicitis [9, 10]. Untreated infection in women can ascend to the upper genital tract and cause salpingitis and lead to pelvic inflammatory disease with scarring and fibrosis of the affected tissues which can result in chronic pelvic pain, ectopic pregnancy and tubal infertility [11]. Other adverse pregnancy outcomes include miscarriage, stillbirth and preterm labour, although studies show conflicting results [12, 13]. Infection in men generally presents as urethritis which can lead to epididymo-

orchitis and possibly infertility [10, 14]. Current Norwegian guidelines recommend genital infections to be treated with doxycyclin 100 mg twice daily for 7 days, alternatively azithromycin one gram single dose can be used if poor compliance is anticipated [15]. As chlamydia antimicrobial assays are complex, non-standardised and difficult to interpret, antibiotic resistance is not routinely assessed in the laboratories [16, 17]. False positive test results may occur up to three weeks after treatment due to persistent DNA [18]. It may be difficult to distinguish between *C. trachomatis* treatment failure and reinfection because of the possibility of re-exposure to an infected partner [5, 19]. Non-compliance should also be considered if test of cure is positive. No vaccine against genital *C. trachomatis* infection is yet available [20].

1.3 Detection and typing

Increased testing for *C. trachomatis* became possible in the 1980's when inefficient cell culture systems were replaced by direct fluorescent microscopic assays, and later by enzyme immunoassays. In the period 1996 to 1999, most Norwegian laboratories implemented the currently used nucleic acid amplification tests (NAATs) that retain both high specificity and sensitivity when applied to urine and vaginal swab specimens [21]. Culture-based techniques are no longer used in Norwegian laboratories. NAATs provide high throughput and are presently the gold standard for chlamydia detection in well resourced settings. The possibility to use first-void urine (FVU) samples in both females and males has expanded testing in non-clinical settings, including high schools.

Genotyping of *C. trachomatis* has a wide range of applications: to examine genetic population structure, as a tool in epidemiologic studies, to reveal transmission in sexual networks, to discriminate between repeat and persistent infections, to detect clonality in an outbreak investigation, and in surveillance of emerging strains such as the Swedish new variant of *C.*

trachomatis (nvCT) [22]. It is assumed that persons infected by the same chlamydia strain are more likely to be epidemiologically linked than those infected with different strains.

Historically, antibodies recognising the major outer membrane protein (MOMP) were used to separate *C. trachomatis* into serovars [23]. *ompA* sequencing is based on the gene encoding MOMP and has been the most widely used typing scheme in *C. trachomatis* in the past decades. It has higher resolution than immunotyping and separates chlamydia into the genovars A-C associated with trachoma, D-K with urogenital infections, and L1-L3 with lymfogranuloma venereum [24]. As the most prevalent genovar E has been detected in about half of chlamydia urogenital infections in heterosexual populations worldwide, recent research has focused on developing genotyping methods with higher discrimination [24-28].

The availability of whole-genome sequencing (WGS) has led to the development of several new genotyping systems. By 2009, four DNA typing methods for *C. trachomatis* genotyping had been published. Two different multilocus sequence typing (MLST) schemes both using 7 housekeeping genes with a resolution similar to that of *ompA* sequencing were available [29, 30]. These are most useful when exploring long term trends in evolutionary studies. In addition, two schemes with higher resolution had been described; a multilocus variable number of tandem repeats (VNTR) analysis published by Pedersen *et al.* in 2008 [31], and an MLST scheme based on 5 highly variable targets of non-housekeeping genes that was developed by Klint *et al.* in Uppsala, Sweden in 2007 [32]. This MLST scheme had been used in several studies in neighbouring country Sweden and was chosen due to high resolution and to enable comparison of sequence types (STs) sampled in the Finnmark High School Study to those included in the Uppsala University MLST database, http://mlstdb.bmc.uu.se. No previous studies had applied high-resolution genotyping in *C. trachomatis* samples from heterosexual persons in Norway or used it as an epidemiologic tool to examine genetic diversity in samples from a general adolescent population including both genders.

1.4 Epidemiology

Chlamydia trachomatis infection is the most commonly reported bacterial sexually transmitted infection (STI) among heterosexual persons in developed countries worldwide [33-35]. True incidence of infection is assumed to be higher than the reported numbers due to its asymptomatic nature. In Western countries, more than two-thirds of all genital chlamydia infections are detected in persons aged 15-24 years, more often in females than males [33, 34]. In 2009, a total of 344 000 cases were reported in Europe, an overall incidence rate (IR) of 185/100 000 [33]. Norway had the third highest chlamydia IR (467/100 000). As 88% of the chlamydia infections in 2009 were reported by four countries (Denmark, Norway, Sweden and the United Kingdom) the results may primarily reflect high levels of testing and thorough reporting in these countries.

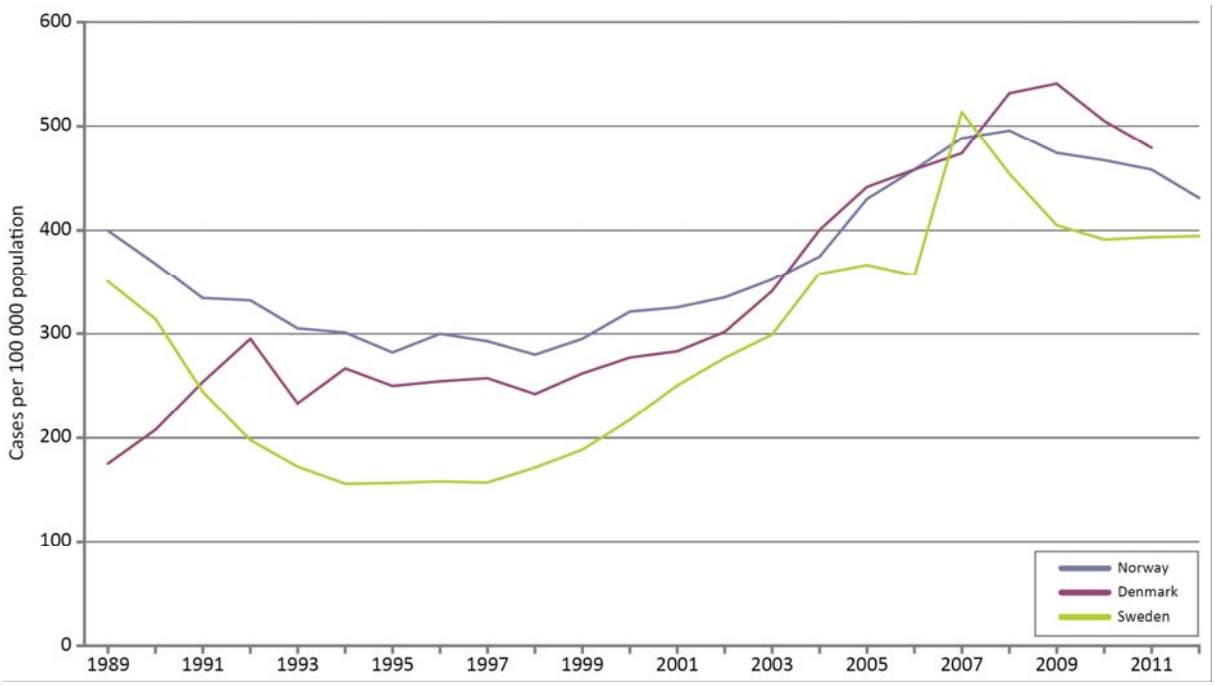

Figure 1. Number of chlamydia cases per 100 000 population reported from the laboratories in Norway, Denmark and Sweden from 1989 to 2012 [36-39].

The chlamydia IRs in Norway have followed a similar pattern as that of Denmark and Sweden; a decreasing trend from the late 1980s to the mid-1990s, followed by a continuous

increase and more than doubling of IR until 2008, while lately a small annual reduction has been observed (Figure 1) [33, 37-39]. The peak in IR in Sweden from 2006 to 2007 was caused by the identification of the mutated nvCT with a deletion in the cryptic plasmid that included the targets for two common commercial diagnostic tests. The nvCT had thus escaped detection in the preceding years [40].

The general increase in chlamydia rates observed in many Western countries since the mid-1990s has been explained by the use of more sensitive diagnostic tests, increase in screening coverage and frequency, improved targeting of risk groups, and possibly a true increase due to changing sexual behaviour [33, 41]. The arrested immunity hypothesis introduced by Brunham *et al.* in 2005 suggests that screening may have increased reinfection rates because early detection and treatment may diminish the immune response [42].

A chlamydia IR almost twice the Norwegian average has been reported in Finnmark, the northernmost county (Figure 2), with an IR of 898/100 000 in 2009 [43].

Figure 2. Map of study area.

In Finnmark, the chlamydia IR has peaked in females aged 15–19 years, while in males the highest IR has been observed among the 20–24 year olds (Table 1). In contrast, the national IR has peaked in age group 20-24 years in both females and males which is in line with surveillance data from Denmark and Sweden [38, 39]. The evaluation of local prevalence data and risk factors to plan chlamydia interventions has been emphasised [44, 45].

Table 1. Numbers of incident chlamydia cases per 100 000 population in age group 15-19 years and 20-24 years by gender in Finnmark county and Norway in 2009 [43].

	Females		**Males**	
	15-19 years	**20-24 years**	**15-19 years**	**20-24 years**
Finnmark	918	584	225	419
Norway	313	412	81	245

1.5 Gender differences in chlamydia prevalence among adolescents

By linguistic definition, *sex* refers to physiological and biological characteristics, while *gender* refers to behaviours, roles, expectations and activities in society [46]. As my thesis examined gender differences in sexual behaviour and testing patterns, the word *gender* is used throughout.

The finding that girls in age group 15-19 years have higher chlamydia IRs than same-aged boys in surveillance data has commonly been explained by more screening opportunities for young women and girls more actively seeking health care [33, 34, 43]. However, a number of cross-sectional studies among adolescents in Western countries show significantly higher chlamydia prevalence in girls than in their male peers, both in school based settings in Southern Norway [47], Luxembourg [48] and the US, [49] and in the general population in

the Netherlands [50], England [51] and Germany [52]. The discrepancy has been linked to cervical ectopy with increased biological susceptibility in adolescent girls [53] and to the possibility that male-to-female transmission may be more efficient than that of female-to-male transmission [54]. In addition, social and cultural factors may contribute [49, 55]. No studies including biological specimens had assessed gender-specific associations between early sexual behaviour and chlamydia infections in age group 15-20 years prior to the Finnmark High School Study. Only a few population-based studies reported prevalence in boys this age [48-50].

1.6 Early sexual behaviour

Adolescence is a period of rapid biological, mental and social development where lifestyle and behaviours with impact on future sexual health frequently are initiated. Sexual behaviour has been shown to vary over time and between cultures and to be deeply rooted in the social or gender constructs of a society [56-60]. The Nordic countries have more liberal attitudes towards female and adolescent sexuality than most other Western countries, and the sexual culture is characterised by equality between genders [61, 62]. However, Nordic data have indicated gender differences in age at first intercourse, number of coital partners, and type and amount of sexual experience [62-67]. Sexual intercourse in adolescents has been accepted provided they feel 'mature enough for sex', which ideally has been associated with being in love with the partner, being in a committed relationship, and acting responsible by using contraception [61, 63]. This 'love ideology' has traditionally been most important for girls [62, 67]. In the Nordic countries, genital intercourse has been introduced early in the stepwise accumulation of sexual experiences following culturally distinct 'sexual scripts' which refers to norms for when, where, and what you can do, and with whom you can have sex [61, 68, 69]. Patterns of early sexual behaviour have converged between genders, and since the early 1970s girls in the Nordic and a few other countries in Northern Europe have experienced first

sexual intercourse earlier than boys [57, 70]. Adolescent Norwegian girls have reported steady couple relationship at a younger age than boys, more frequent and regular sexual intercourse and older sexual partners [65, 66], while boys have had more varied sexual experience including casual sex and multiple partners [62, 63, 70]. In 2002, median sexual debut age among Norwegian girls was 16.7 years and in boys 18.0 years, a decrease from 17.7 and 18.5 years, respectively, since 1992 [67]. In 2011, a Nordic study reported a median age at first intercourse of 16 years among Norwegian girls indicating a further decrease [71]. A Norwegian study from 2003 found that adolescent females were as inclined as males to break the norm of being in love as the basis for a sexual relationship [67]. With a majority of girls preoccupied with older partners, adolescent boys can either enter a relationship with a younger girl not 'feeling mature enough for sex', or rely on multiple occasional relationships and sporadic sex, and lower coital frequency due to less access [62]. The liberated attitude towards female sexuality, combined with the average girl entering puberty at a younger age than the average boy, girls dating older partners, and having easy access to oral contraception can explain why sexual activity over the past decades has been initiated and peaked earlier in girls than boys in the Nordic countries.

1.7 Chlamydia surveillance

Most European countries report some system for surveillance of genital chlamydia infections [72]. Until 2002, surveillance of chlamydia infections in Norway was based on voluntary aggregate reporting from all laboratories to the Norwegian Institute of Public Health (NIPH) [21]. In 2003, genital chlamydia infections became mandatory notifiable and part of the Norwegian Surveillance System of Communicable Diseases. Since 2005, the laboratories are required to report year of birth, gender, municipality of residence, and localisation of infection to NIPH. Our understanding of the chlamydia epidemiology in Norway is largely based on

surveillance data. As these data lack unique individual identifiers, accurate annual testing rates and repeat testing rates in the general population cannot be estimated.

1.8 Testing and screening

Testing is a crucial part of any chlamydia control strategy. With mostly asymptomatic chlamydia infections, a high proportion of those infected have no physical clue to seek health care [8]. Screening is defined as testing for chlamydia to detect and treat infections in people who do not necessarily perceive themselves to be at risk or do not know if they are infected, with the intention to reduce future morbidity [72]. Two distinct screening approaches exist. Opportunistic screening implies a health professional offering a test to patients attending health care for any reason with the health professional responsible for repeating the test offer at regular intervals. Systematic screening uses registers to identify, invite and remind the target population to be tested irrespective of health service use. The screening frequency and coverage required to reduce chlamydia prevalence and its complications remains unknown [73].

In Norway, there is no official screening programme. The Norwegian guidelines recommend testing of both females and males in the presence of clinical symptoms, or if partner is infected, or in persons aged < 25 years after change of sexual partner, or in women presenting for termination of pregnancy or antenatal care [15, 74]. According to law, testing and treatment in these groups is free of charge [75]. Test of cure 5-6 weeks after treatment and notification to sexual partners over the past 6 months is recommended [15, 74].

Chlamydia testing of adolescents is widely available in Norway. The majority of testing is done in general practice and in public youth clinics which are tailored to the needs of adolescents and are present in most municipalities [76]. Youth clinics provide contraceptive counselling without parental consent and all services are free of charge. School based

chlamydia screening is not current policy in Norway. Most high schools have a school nurse available part time offering limited STI testing as part of the general health service. Specialist STI clinics are present only in large Norwegian towns. Hospital outpatient clinics in venereology and gynaecology only accept referred patients.

In Western countries having implemented chlamydia control strategies, young females are tested far more frequently than young males [33, 38, 39, 51, 77]. According to annual Norwegian surveillance data 2007-11, the average female to male chlamydia test ratio in age group 15-19 years was 4.5 to 1, and in age group 20-24 it was 2.8 to 1 [43].

More adolescent girls and particularly boys are reached if chlamydia testing is extended to high schools and other non-clinical settings [78-80]. Extensive high school based chlamydia screening and treatment programmes including both genders have been conducted in the US; in Philadelphia [49], New Orleans [81], New York [82] and San Francisco [44], with participation ranging from 52% to 65%. European high school based screenings have reported 63% participation in Southern Norway [47], 38% in Luxembourg [48], and 73% in a small vocational school study in the Netherlands [83]. A recent systematic review of chlamydia screening in educational settings found that classroom based approaches achieved the highest test rates [84]. None of the previous school based screenings had examined the behavioural factors associated with being chlamydia tested at school.

2. Aims of the thesis

The overall aims of the thesis were to examine multiple aspects of the high chlamydia IRs among adolescent girls and boys in Finnmark, including chlamydia genotypes, disease prevalence, early sexual behaviour, and factors associated with testing. The specific aims for each paper were:

I. To examine the distribution of *C. trachomatis* genotypes in a general adolescent population in a rural high-incidence area in Norway, to compare chlamydia genetic diversity in this area with that of two urban regions, and to compare discriminatory capacity of two different genotyping methods; multilocus sequence typing and *ompaA* sequencing.

II. To detect chlamydia prevalence in adolescents aged 15-20 years in a high-incidence area in Norway, and to examine gender-specific early sexual behaviours associated with chlamydia infections.

III. To determine the proportions of adolescents tested in a high school based screening and previously in clinical practice, to detect chlamydia prevalence according to testing pattern, and to examine demographic and sexual behaviour characteristics associated with testing.

3. Materials and methods

3.1 Study population

The Finnmark High School Study (FHSS) was conducted as a population based cross-sectional study in 5 public high schools in Finnmark county, Norway. Finnmark has a sparse population living in minor municipalities and borders Northwest Russia to the east, Finland to the south and east, and Troms county to the west (Figure 2). By sea, it borders the Atlantic Ocean and the Barents Sea. The population includes ethnic Norwegians, indigenous Sami people, and minority groups of Kvens, Finns and Russians. Data were collected during 9 weeks from September to November 2009 using web-questionnaires and first-void urine (FVU) samples. The principal in each school consented to participation. All 1,908 students in the high schools in the coastal municipalities Hammerfest, Kirkenes, and Alta, and in the inland Sami municipalities Karasjok and Kautokeino were invited. The student lists for each class were the basis for the invitations. All data were collected by the same experienced female doctor (principal investigator) and nurse who consecutively visited a total of 123 classes using an identical classroom based approach. In each municipality, the study staff gave tailored lectures on logistics, sexual behaviour and chlamydia infections to principals, teachers, school nurses, general practitioners and youth clinic staff prior to data collection. An invitation letter with information about chlamydia infection, questionnaire items, sampling procedures, and use of data in both Norwegian and Sami was handed out in class two weeks before data collection (Appendices 1-3). Confidentiality regarding questionnaire data and chlamydia test results was assured both in the written information and repeated orally in each class on the day of data collection. The students were informed about the mostly asymptomatic nature of genital chlamydia infections and the value of testing to prevent adverse health outcomes. The high chlamydia rates among adolescents in Finnmark were

emphasised. Chlamydia testing was promoted as a good and responsible thing to do. The testing equipment was displayed in class prior to urine sampling. Overall participation rate was 85% (1,618 of 1,908 invited students provided questionnaires and/or urine samples).

3.1.1 Inclusion and exclusion criteria

Paper I: The genotyping study included 60 chlamydia specimens from 1,476 urine samples with a valid chlamydia test result collected from participants in the FHSS. Parallel to the FHSS, 20 and 80 chlamydia test positive urine samples from 15-20 year olds in Finnmark and Tromsø, respectively, were consecutively collected from routine clinical samples in the laboratory of the University Hospital of North Norway (UNN Tromsø) (Figure 3). 88 samples from the same age group in Trondheim were collected at St. Olavs Hospital in Trondheim, Central Norway. Thus, a total of 248 chlamydia samples were available for genotyping.

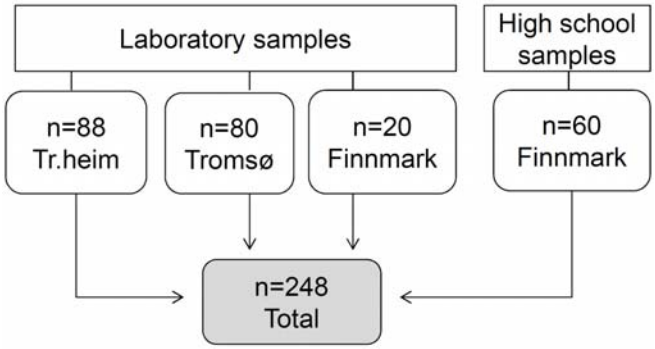

Figure 3. Chlamydia urine samples, Paper I.

In a separate analysis, we calculated mean age of last sexual partner in the 1,031 high school study participants with valid questionnaire and urine sample reporting sexual intercourse (Figure 4).

Paper II: The study population in the paper on early sexual behaviour and chlamydia infection is shown in Figure 4. If only assessing students present at school and thus eligible, 2% (46 of 1,664) refused participation. 442 participants responding ‘no’ to: ‘Have you ever had sexual

intercourse?' were considered not to be at risk for chlamydia infection and were excluded from the analyses. All 442 had negative test results. Among 6 students with inconclusive test result, one girl testing negative one day prior to data collection was assumed to be negative and was included in the analysis. 5 boys with an inconclusive test result did not provide a new sample when asked and were excluded. A total of 1,031 participants aged 15–20 years with sexual intercourse experience, questionnaires and valid chlamydia test results were included in the study. 59 of these had a positive chlamydia urine sample.

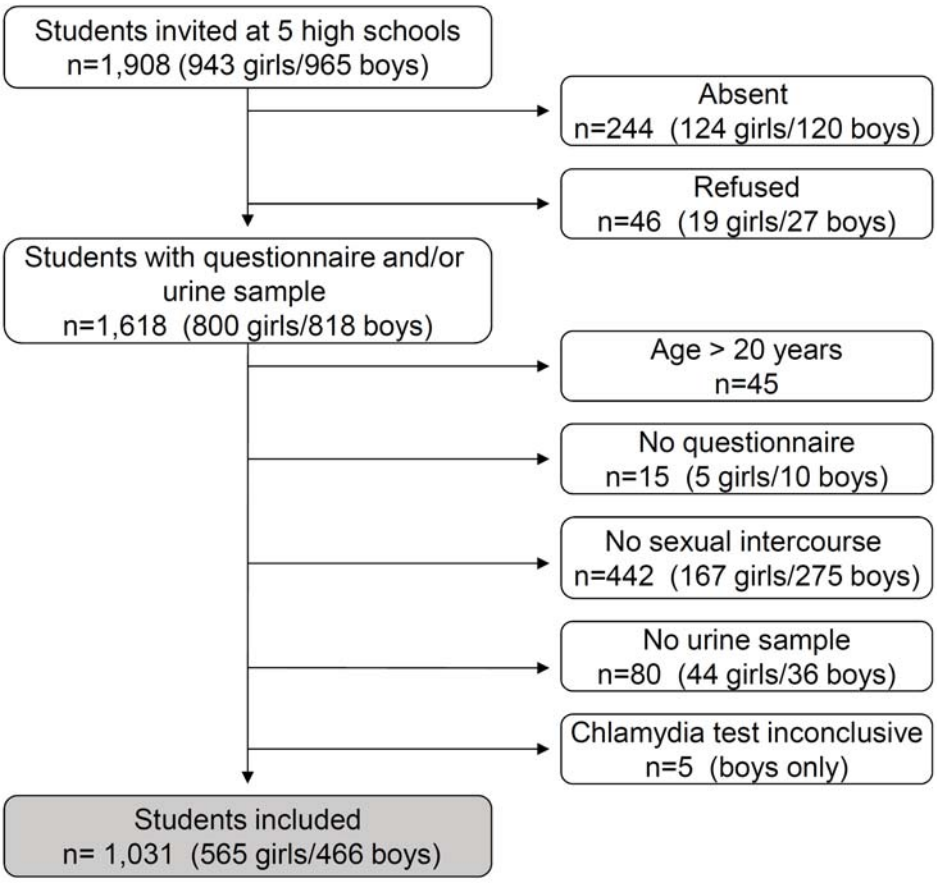

Figure 4. Study population, Paper II.

Paper III: In the study of chlamydia testing, a total of 1,112 participants aged 15-20 years with questionnaires that included valid response to previous chlamydia testing, and with sexual intercourse experience were included in the analysis (Figure 5).

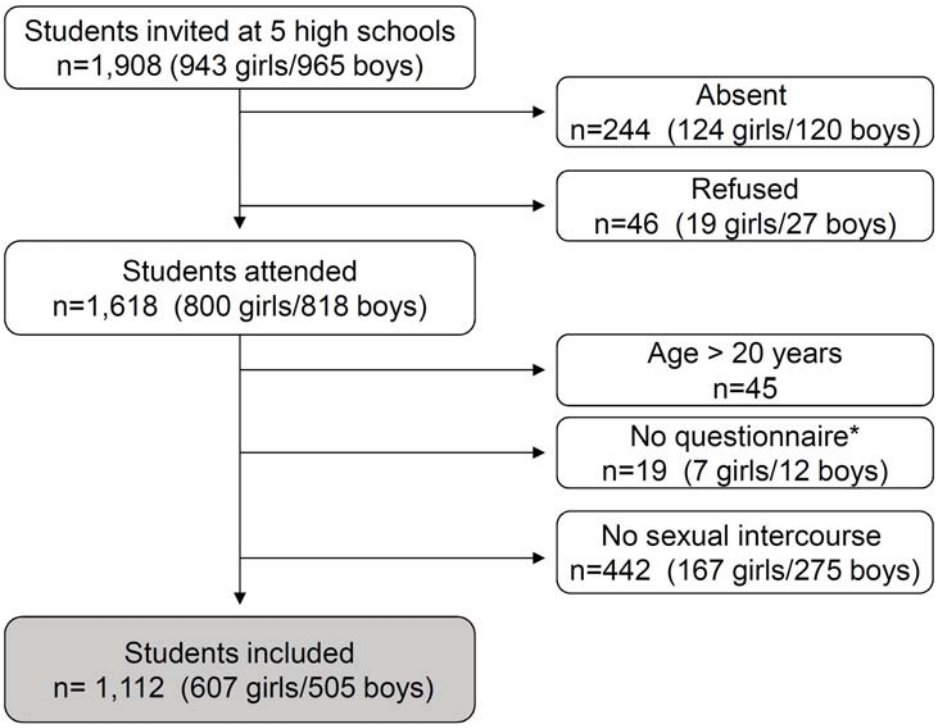

Figure 5. Study population, Paper III. *Missing questionnaire (n=15) or missing response to the question 'previous test' (n=4).

3.2 Sample size calculations (Paper II)

We estimated a sample size of 974 to achieve 90% power to detect a difference between an anticipated chlamydia prevalence of 3.0% in the source population irrespective of sexual intercourse experience, compared to 1.4% as observed in a similar study in Southern Norway using a 5% significance level [47]. The anticipated prevalence was based on a pilot study in April 2009 in Lakselv high school in Finnmark (unpublished data).

3.3 Questionnaire

The questionnaire was developed for the FHSS and contained a total of 68 questions of which one third had sub items (Appendix 4 and 5). It included validated questions used in 5 nationwide surveys of sexual behaviour in 1987, 1992 and 2008 (age group 18-60 years), and 1997 and 2002 (18-49 years) [64], in a national survey of sexual behaviour in age group 17-19 years in 1989 [62], as well as in a prospective survey of adolescent sexual behaviour in Nordland County in Norway from 1999 to 2001 [85]. Ethnicity and religious affiliation was assessed using questions from the North Norwegian Youth Study 1994-95 [86]. The FHSS questionnaire was designed in QuestBack online survey system (www.questback.com) and was tested for comprehensibility, clarity and time use in a pilot study including 31 students in Lakselv High School, year 1-3, in April 2009. It was adjusted according to feedback from the participants.

On the day of data collection, the questionnaire was emailed class-wise to the students 10 minutes before the study staff arrived in the classroom. All Norwegian high school students manage their own laptop computers with internet access making this approach feasible. Under supervision of the study staff and a teacher, participants spent 10-20 minutes completing the questionnaire which included questions on demography, substance use, sexual behaviour, contraceptive use, current urogenital symptoms, and earlier chlamydia testing and treatment (Appendix 4 and 5). Pre-programmed commands ensured automatic skipping of non-applicable questions. Persons with no sexual intercourse experience answered alternative questions on attitudes and feelings towards sex, intimate non-coital experiences, and STI knowledge ensuring that time spent on the questionnaire was independent of sexual experience. No reminders were sent.

3.3.1 Data from questionnaires

Self-perceived ethnicity was coded in three categories based on the statement: ’I perceive my ethnicity as: Norwegian, Sami, Russian, Kven, Finnish, or other’. Kvens are descendants of Finnish-speaking immigrants from Northern Finland and Sweden [86]. More than one answer was allowed. Category ’Norwegian’ included those reporting Norwegian (n=726) and/or Kven (n=5) ethnicity, as the two share a common distribution of lifestyle factors [87]. ‘Sami/ Sami-Norwegian’ included those reporting Sami ethnicity (n=90) or Sami and Norwegian ethnicity (n=139). ‘Other’ included Russian (n=19), Finn (n=20) and other (n=31) ethnicity.

Participants’ residence during school year was reported as: 1) At home with my parents, 2) Living with grandparents/other relatives, 3) Private room/apartment, 4) Student house, 5) Host family, or 6) Other. Due to small groups, the variable ‘Residence during school year’ was dichotomised as: ‘At home with my parents’ (response 1) and ‘Other’ (responses 2-6).

The variable ‘high school study affiliation’ was defined as; 1) ‘academic’, including students in the general academic studies programme, and 2) ‘vocational’, including vocational school students. In Norway, academic and vocational classes frequently share facilities throughout high school.

Use of alcohol, cannabis, amphetamine or ecstasy was reported for each substance as: never tried (1), tried (2), occasional use (3), or regular use (4). A new variable ‘alcohol/drug use’ was calculated as sum of the four substance use variables. Participants with missing response for alcohol (n=5) were excluded, but this was accepted for the other three. Range of the ‘alcohol/drug use’ variable was 2–16, and was defined as: $\leq$5: ‘low’; 6: ‘medium’; $\geq$7: ‘high’.

Young age at first intercourse was defined as $\leq$14 years in accordance with a recent study assessing risk-taking behaviours among Nordic women [88].

Condom use at first intercourse with first partner and at last intercourse with last partner were coded in two categories (yes/no) based on the question: 'Did you use any contraception at first (last) sexual intercourse?' with response alternatives: 1) No, 2) Condom, 3) Hormonal contraception, 4) IUD, 5) Both condom and other contraception, 6) Emergency pill, 7) Coitus interruptus, 8) Don't know. Category 'yes' included participants with response 2) or 5). 'No' included the remaining responses. 'Don't know' was answered by 3 girls and 10 boys at first intercourse, and by 3 girls and 8 boys at last intercourse.

Previous clinic based testing was assessed by; 'Have you previously been tested for genital chlamydia infection?' with response options: 'Yes, once', 'Yes, twice', 'Yes, 3 times', 'Yes ≥4 times', or 'No'. Due to small groups, the variable 'clinic based testing' was dichotomised as yes/no. We assumed all previous testing to be clinic based, i.e. youth clinics and general practice and only occasionally in STI clinics and hospital outpatient clinics. 'School based screening' included all participants that were screened in the high school study independent of clinic based testing. The subgroup 'school-only test' included participants with no previous clinic based testing that provided a urine sample in the school based screening.

3.4 Collection of urine samples

After finishing the questionnaire in the classroom, participants went directly on to the school toilets where they were instructed how to provide a first-void urine (FVU) sample by the study nurse. Each participant received a test kit that included: 1) a completed laboratory form including three adhesive labels with name, birth date, and mobile phone number, 2) a urine collection cup with an ink mark at 12 ml, 3) a urine sample transport tube, and 4) disposable gloves. The nurse collected the urine transport tubes immediately outside the toilet and ensured that each person approved the printed personal information on the form and on the transport tube label. The urine samples were refrigerated and transported by National Mail

Delivery on the same afternoon to UNN Tromsø and analysed within 24 hours. UNN Tromsø is the only laboratory for microbiology diagnostic services in Finnmark.

3.5 Follow up

Participants testing positive or inconclusive for chlamydia were phoned by the study nurse on the same afternoon as the laboratory reported the test result. After repeat calls, all were eventually reached and given an appointment at the local youth clinic. All the 60 test positive participants either got a prescription of a single dose one gram azithromycin or were given antibiotics directly for observed treatment. The youth clinic notified, tested and treated sexual partners. All study participants were included in a lottery with three persons winning a mobile phone with a one year subscription worth 140 Euros in 2009.

3.6 Laboratory testing

3.6.1 Chlamydia PCR

The UNN laboratory extracted DNA using the BUGS'n BEADS TM-STI kit (NorDiag ASA, Oslo, Norway) and used ProCt real-time PCR (ProCelo A/S, Tromsø, Norway) with sensitivity 97% and specificity 100% (*incA* gene and internal control). The St. Olavs Hospital's laboratory prepared DNA using the bacterial protocol on GenoM (Qiagen, Hilden, Germany) and used an in-house triplex real-time PCR (cryptic plasmid, MOMP gene and internal control) with sensitivity 96% and specificity 100% [89]. A plasmid specific PCR was used to confirm MLST identification of nvCT [40].

3.6.2 Chlamydia trachomatis genotyping

All the 248 chlamydia samples were immediately frozen at -70°C in the laboratories and later transported on dry ice to the University Hospital of Uppsala, Sweden, for genotyping. *ompA* sequence determination was performed according to a previously described method [90]. Strains were categorised into genovars D-K and *ompA* genotypes. Genovars are *C.*

trachomatis subgroups based on serospecificity for MOMP inferred from *ompA* sequencing. Genotypes are subgroups based on *ompA* sequencing. The MLST scheme comprising 5 highly variable target regions was performed according to Klint *et al.* [32] and modified according to Jurstrand *et al.* [91]. Each sequence type (ST) is based on 5 digit strings that represent the different alleles. Allele profile numbers were assigned by comparing the sequence at each locus to the Uppsala University *C. trachomatis* database (http://mlstdb.bmc.uu.se). New allele numbers were assigned in order of discovery. Clonal complexes were defined as clusters of STs with only one allele difference, i.e. single locus variants (SLVs). The founder of a clonal complex was the ST with the highest number of SLVs.

3.7 Statistical methods

Paper I

The discriminatory power (*D*) of a typing method, the probability that two unrelated strains sampled randomly from a test population will be categorised in different groups, was calculated for *ompA* genotyping and MLST in the 188 routine clinical samples. We used Hunter and Gaston's modification of Simpson's discriminatory index [92]:

$D = 1 - \frac{1}{N(N-1)} \sum_{j=1}^{S} n_j (n_j - 1)$, where N is the total number of strains tested, s is the total number of different types, and n_j is the number of strains belonging to the jth type. Confidence interval (CI) for *D* was calculated as originally described by Simpson [93]. The guidelines outlined by the European Society of Clinical Microbiology and Infectious Diseases (ESCMID) state that a molecular typing method should have a *D* of ≥0.95 to be considered 'ideal' [94]. As the 188 samples were consecutively collected in the laboratories from a defined age group within a limited time frame from defined geographic areas, some epidemiological dependence could not be excluded. In order to adjust for this possible dependence, the following corrections were made: the two most common STs were assumed

to have prevalence equal to the third most prevalent ST. Thus, the number of strains in ST12 (n=35) and ST56 (n=28) were set equal to the number of strains in ST153 (n=15), and a corrected D_c was calculated.

Minimum spanning trees were generated by an analysis of the full MLST profiles in all 248 specimens using BioNumerics software (version 6.01, Applied Maths, Sint-Martens-Latem, Belgium) under the categorical coefficient of similarity and the priority rule of the highest number of SLVs.

Chi-square test was used to assess associations between urogenital symptoms and STs and clonal complexes. The 95% CI for proportions was calculated using Clopper-Pearsson's exact method.

Paper II and III

Descriptive characteristics were reported with means (standard deviation) for continuous variables and with numbers (%) for categorical variables. The 95% CI for proportions were calculated using the exact binominal method. Crude and multivariable logistic regression models were applied using chlamydia test result (positive/negative) as the outcome variable in Paper II. In Paper III, two outcome variables were used: 1) clinic based testing, i.e. if participants had been chlamydia tested before the FHSS (yes/no), 2) school based screening, i.e. if participants were tested in the FHSS (yes/no). Variables with p value <0.25 in crude analysis were included in the multivariable regression models which were fitted using stepwise procedures. Age and gender (if applicable) were included regardless of significance. Collinearity was not a problem with variance inflation factor (VIF) <2.5 for all variables. Gender interaction was assessed by including cross-product terms between each independent variable and gender. All statistical tests were two-sided using a 5% significance level. SPSS version 18.0 (Paper I) and SPSS 19.0 (Paper II and III) were used for all statistical analyses.

In Paper III, one statistically significant interaction term was included in the final multivariable model, and model fit was assessed using Hosmer and Lemeshow goodness-of-fit test with 5 of 6 *p* values >0.25.

3.8 Ethics

In the FHSS, written informed consent was obtained from the next of kin, carers or guardians on the behalf of participants younger than 16 years. Participants ≥16 years gave their informed consent by filling in the web-based questionnaire in accordance with the Health Research Act §17.b stating their right to consent. All procedures were approved by the Regional Committee for Medical and Health Research Ethics North Norway (REC North No.: 200900528-6/MRO/400) and the Data Protection Officer at UNN (Number 2009/2475). Establishment of a research bio-bank for *C. trachomatis* urine samples was approved by The Norwegian Directorate of Health (Bio-bank Registry Number 2723).

4. Summary of results

Paper I: Multilocus sequence typing of genital Chlamydia trachomatis in Norway reveals multiple new sequence types and a large genetic diversity

In 248 specimens from the previously unmapped areas Finnmark, Tromsø and Trondheim, *ompA* sequencing detected 11 genotypes while MLST displayed 50 sequence types (STs) thus providing 4.5 higher resolution. A total of 12 alleles in the MLST scheme and two-thirds of all STs were novel. The common genovar E comprised 46% of all specimens and resolved into 24 different STs. MLST identified the new Swedish variant of *C. trachomatis* not discriminated by *ompA* sequencing in 1.6% of samples. Simpson's discriminatory index, *D*, for MLST was 0.93 (95% CI 0.91-0.95), while the corrected index, D_c, was 0.97 (0.96-0.98). For *ompA* sequencing, *D* was 0.67 (0.61-0.73). There were no statistically significant differences in genetic diversity of STs between the three areas. Finnmark had an atypical genovar distribution with G being predominant, mainly due to the expansion of ST128 and the novel ST161. The latter was unique for Finnmark.

Paper II: Early sexual behaviour and prevalent Chlamydia trachomatis infection

Prevalence of chlamydia infection was 5.7% (95% CI 4.4-7.3%). Girls were twice as likely to be infected as boys, 7.3% (5.3-9.7) versus 3.9% (2.3-6.0). Girls reported significantly earlier sexual debut, older sexual partners, more steady relationships, higher lifetime number of sexual partners, and less condom use at last sexual intercourse than boys. Boys reported higher levels of substance use overall and in connection with last intercourse. In girls, higher maternal education (odds ratio, OR, 2.22, 95% CI 1.13-4.37), $\geq$ 2 sexual partners past 6 months (OR 3.59, 1.76-7.32), and partner meeting venue at a private party, bar or disco (OR 4.99, 1.10-22.69) increased the odds of infection in the multivariable model. In boys, condom use at first intercourse (OR 0.06, 0.01-0.42) decreased the odds of infection, while having an older last sexual partner (OR 3.74, 1.27-11.01) increased the odds. In girls and boys

combined, the risk of infection increased if residing outside the family home during the school year (OR 2.04, 1.17-3.57), ≥ 2 partners past 6 months (OR 2.88, 1.60-5.18), and meeting last sexual partner at a party, bar or disco (OR 3.54, 1.18-10.61), and decreased if condom was used at last intercourse (OR 0.23, 0.07–0.75). In Table 3, the correct values for 'meeting last partner on the Internet' for girls and boys combined should be OR 2.81 (0.78-10.08).

Paper III: Factors associated with Chlamydia trachomatis testing in a high school based screening and previously in clinical practice

56% of girls and 21% of boys reported previous clinic based testing. In the school based screening, 93% were tested with no gender difference. 42% of girls and 74% of boys were tested for the first time at school ('school-only test'). Both girls with clinic based testing and girls with school-only test had high chlamydia prevalence (7.3% vs 7.2%). Boys with clinic based testing had twice the prevalence of boys with school-only test (6.2% vs 3.0%, p=0.01). Half of infections were detected in participants with school-only test. One-fifth were repeat infections. In multivariable analysis of girls and boys combined, the following variables increased the odds of clinic based testing: older age (OR per year 1.54, 95% CI 1.30-1.83), first intercourse ≤14 years (OR 2.02, 1.43-2.85), no condom use at first intercourse (OR 1.48, 1.09-2.01), steady relationship (OR 1.51, 1.11-2.01), and higher number of lifetime partners: 1-2 partners (reference), 3-5 (OR 3.07, 2.11- 4.46), and ≥6 (OR 7.63, 5.03-11.55). Significant interaction was present between gender and ethnicity (p=0.012). In all ethnic groups, females had higher odds of previous test than males (females versus males): Norwegian (OR 7.96, 5.26-12.04), Sami/Sami-Norwegian (3.62, 1.92-6.82) and other (OR 1.89, 0.66-5.45). In the multivariable analysis with school based screening as outcome variable, the following variables decreased the odds: female gender (OR 0.57, 0.34-0.97), vocational affiliation (OR 0.51, 0.30-0.87), first intercourse ≤14 years (OR 0.58, 0.35-0.95), and no condom use at first intercourse (OR 0.57, 0.35-0.94). In addition, current urogenital symptoms (OR 3.23, 1.57-6.65) increased the odds of school based screening.

5. Discussion

The FHSS was the first high school based chlamydia screening in Europe to include both girls and boys in all three year levels and to use a comprehensive questionnaire to assess sexual behaviour. Our study was unique in applying an interdisciplinary approach that included public health aspects, mapping of sexual behaviours and testing patterns, detection of chlamydia in high quality biological samples, and the use of an advanced high-resolution method to genotype *C. trachomatis*. The FHSS was limited by cross-sectional design and self-reported questionnaire data. The study had low statistical power to assess associations between demographic and sexual behaviour factors and chlamydia infection due to the small number of chlamydia positive urine samples.

5.1 Internal validity

An internally valid effect is one that correctly describes the association between exposure and outcome in the target population. Three types of systematic errors may threaten the internal validity: (i) selection bias; distortions resulting from procedures used to select subjects and from factors that influence study participation, (ii) information bias; different consequence of errors in measurement of exposure and/or disease in subjects, and (iii) confounding factors; the extraneous factors responsible for difference in disease frequency between exposed and unexposed.

5.1.1 Selection bias

Selection bias occurs if there are systematic differences in the exposure status between participants and non-participants in the study. High participation may reduce the potential for selection bias. In Finnmark county, 94% of the birth cohort was enrolled in high school from 2007-09 with an annual drop-out rate of approximately 10% [95]. An estimated number of 167 persons were lost due to drop-out throughout high school and were thus not included

(calculations not shown). Studies differ as to whether drop-outs are at increased STI risk [96, 97]. We may have underestimated levels of risk behaviours and chlamydia prevalence if drop-outs and other non-attendees had higher prevalence than the high school students.

Among non-attendees, 244 students were absent from school when the study was conducted due to excursions, field work, job training, disease or other reasons and were thus not eligible (Figure 4). Only 2% (46 of 1,664) of eligible students refused participation for unknown reasons, thus limiting the potential for selection bias [98].

5.1.2 Information bias

Information bias refers to bias related to instruments and techniques used to collect information about exposure and outcome variables [99]. Differential misclassification may occur if the misclassification of exposure is associated with outcome status. The high school study participants did not know their chlamydia test result when filling in the questionnaire. Differential misclassification is possible, but we had no reason to believe there was a high level of this bias. Non-differential misclassification may occur when all categories of a variable (exposure, outcome or covariate) have the same probability of being misclassified for all participants.

The urine sampling and labelling procedures in the FHSS ensured correct linking between the persons' identity and the urine sample for each participant. Exchange of urine samples between participants is unlikely due to thorough supervision.

False positive chlamydia test results in the 60 high school samples (Paper I, II, III) and in the 188 clinical routine samples (Paper I) are unlikely as all 248 *C. trachomatis* specimens were successfully genotyped using *ompA* sequencing and MLST, providing evidence for presence of chlamydia DNA in these samples. DNA contamination in the genotyping laboratory in Uppsala is unlikely due to the finding of multiple different STs (52% of STs comprised only

one specimen and 62% had <4 specimens). Batches of specimens from all three geographic areas were analysed simultaneously, and the finding of the unique ST161 in multiple samples from Finnmark also suggests no contamination.

False negative chlamydia test results in girls are possible because self-sampled vaginal swabs in females have about 10% increased sensitivity compared to FVU samples [100]. The true chlamydia prevalence in girls may thus be higher than the estimated 7.3% (41 cases among 565 girls) with approximately four chlamydia infections remaining undetected giving a prevalence of 8.0% (45 of 565). Furthermore, the reported 97% sensitivity for the PCR test at UNN Tromsø may indicate that we have missed approximately two chlamydia cases in the FHSS. False negative test results in both genders could be caused by sampling error, transport conditions, low bacterial load, laboratory error, and PCR inhibitors. To avoid false negative test results, both the laboratories in Tromsø and Trondheim used an internal amplification control, and positive and negative controls were used both for extraction and setting up the PCR.

Obtaining a complete MLST profile in all 248 specimens was unusual because it implied every single allele PCR returning a valid result (Paper I). The result is plausible because we only used high-quality specimens that were frozen immediately after the initial diagnostic PCR. Additionally, the MLST scheme had been optimised since 2007 and analyses were performed and supervised by an experienced laboratory scientist who was very familiar with the method.

Accuracy of retrospective self-reported data depends on the participants' ability to recall past behaviours and their willingness to report them [99, 101]. Recall bias refers to differences in the accuracy of the recollections retrieved by participants [99]. All our data were self-reported, except for school year, study affiliation, chlamydia prevalence and genotyping

results. To minimise recall bias, the questionnaire proceeded chronologically from sexual debut to the most recent sexual intercourse enabling the respondents to sequentially order recall of events and promote thoughtful response (Appendix 4 and 5) [101, 102]. Personal experiences with high emotional impact such as the question on first sexual intercourse can produce 'flashbulb' memories and may be reported with high accuracy [102]. Single-event recall like last sexual intercourse has shown to be valid representation of sexual behaviours over longer time periods and was used in the questionnaire [103]. A number of sexual behaviour questions were linked to the first and most recent sexual partner because the 'by-partner' approach provides a context and a focus for past events with the potential to reduce recall bias [104].

The use of laboratory data to assess the outcome variable 'clinic based testing' instead of a questionnaire would have eliminated recall bias for this variable. However, recalling autobiographic events is easier if memory contains few similar events such as chlamydia testing [102]. Some recall bias in retrospective reports of sensitive behaviours is to be expected [104]. If recall bias were similar among the infected and non-infected participants, the resulting information bias will be non-differential.

Social desirability bias refers to over-reporting of socially desirable behaviours and under-reporting of undesirable behaviours thus aiming for positive evaluation by others, protecting ones' self-image, and conforming to cultural norms [98, 101]. Our questionnaire included multiple potentially sensitive topics. We therefore used several techniques to reduce social desirability bias that included; i) priming participants' motivation to be honest, ii) computer based self-administration of the questionnaire, iii) confidentiality assurances, and iv) careful wording of the questions [98, 105].

i) The oral information emphasised that high quality in research depends on full honesty in reporting. We further stressed that accuracy in reporting would provide policy makers with valuable data to develop STI programmes that might benefit their age group.

ii) Adolescents may have higher acceptability for answering sensitive questions in a computer-assisted self-interview (CASI) than in a face-to-face interview [104, 106-108]. CASI may also increase accuracy in reporting and motivation to complete the survey. The question assessing number of sexual partners seems to be sensitive in different directions between genders with women under-reporting and men exaggerating, but use of CASI has been shown to reduce such gender disparities in reporting [108, 109]. In Paper II, girls and boys reported similar numbers of sexual partners past 6 months. Significantly more boys than girls reported 1-2 lifetime number of partners (48% vs 35%, $p<0.001$), while more girls than boys reported $\geq$6 lifetime partners (34% vs 25%, $p=0.003$). It is likely that some gender-related social desirability bias was present in the sexual behaviour questions. However, we assume it to be smaller than in studies from Southern Europe and the US due to more liberated attitudes towards adolescent and female sexuality in Norway and to the use of CASI. Furthermore, we observed an expected association between number of sexual partners and chlamydia infection in both girls and boys suggesting high validity of the data.

iii) Confidentiality towards parents, teachers and researchers regarding data handling, storage and analysis was assured both in the oral and written information (Appendices 1-3). We stressed that time spent on the CASI was independent of sexual experience due to design and skipping patterns. The following measures were implemented to increase levels of perceived confidentiality in the classroom during the survey: space between students, use of a CASI with small font readable only at close range, and the presence of three adults. High and universal participation with few missing responses indicates high level of perceived

confidentiality [98]. Accuracy in reporting was indicated by few extreme values in numerical variables and high level of consistency between variables.

iv) Accurate reporting of sensitive behaviours may increase by having a long introduction to the question and thus deliberately loading it [101]. The question assessing reasons for having first intercourse was phrased as follows: 'There are usually many different reasons for a person to have sexual intercourse. What was your reason to have your first intercourse?' The words 'usually' and 'many different reasons' may decrease the significance of the behaviour and increase the respondent's willingness to report on it.

Item response rate was high throughout the questionnaire. Sexual behaviour topics did not suffer from low response rates with 99% answering the question on same-sex experiences and 97% replying to the question on first sexual intercourse. The detection of chlamydia infection only in participants reporting sexual intercourse suggests truthfulness in reporting. This contrasts studies in the US where detection of STIs in adolescents claiming no sexual intercourse experience is common [82, 110].

5.1.3 Confounding

A confounder is defined as a factor that blurs the observed effect and is associated both with the exposure and the outcome [99]. In contrast to selection and information bias, measured confounders can be controlled for in the statistical analysis. All multivariable analyses in Paper II and III were adjusted for gender and age. Gender interaction was assessed for all variables in crude and multivariable analyses, and significant interaction terms were included in the models. Confounding by unknown factors such as the number and timing of concurrent partnerships could not be ruled out [111-113].

5.2 External validity

External validity pertains to the ability to generalise the findings in the study to the general population [99]. To our knowledge, no population based study is currently available for comparison of the ST distribution in the 60 high school urine samples as the MLST scheme so far only has been used in chlamydia samples from patients attending clinical settings (Paper I) [114, 115]. When assessing all 248 samples, the findings that the putative founders of clonal complexes already were present in the MLST database and that a majority of samples belonged to clonal complexes, correspond to MLST databases for other bacteria and thus suggest high external validity [116]. Furthermore, the low prevalence of nvCT corresponds to the limited spread observed in Southern Norway and in other countries also indicating high external validity [117, 118]. Genovar E comprising 46% of all 248 samples resembles other studies on genovar distribution in heterosexual populations worldwide, and thus supports the generalisability of our results [24-28, 114].

The CASI included validated questions that had been used in comparable populations [62, 64, 67, 85, 86]. As the FHSS was confined to a chlamydia high incidence area, we assume levels of sexual risk behaviour and chlamydia prevalence to be higher than in the general adolescent population in Norway (Paper II) [47]. However, the observed gender differences in adolescent sexual behaviour are similar to reported results in other Nordic studies [62, 63]. The majority of risk factors associated with prevalent chlamydia infection (Paper II) correspond to those observed in other high school studies reporting on sexual behaviours [47, 48, 82]. To our knowledge, the association between higher maternal education and chlamydia infection in girls has not been observed in other studies. We found no previous studies assessing the association between residence outside the family home during high school and prevalent infection. Education and residence will be further discussed in the next section. The gender difference in chlamydia prevalence resembles that observed in population based studies

among adolescents in Northern Europe [47, 48, 50]. The external validity of FHSS would have increased if high school students from other parts of Norway had been included.

5.3 Discussion of main results

Systematic errors such as selection bias, information bias or confounding are not likely to explain our main findings in the FHSS. In statistical models, real associations can be missed because of low statistical power, and reported associations may be spurious due to multiple statistical tests. It is likely that we did not detect some real associations due to limited statistical power. In Paper II with 59 chlamydia cases in 1,031 participants, we had 80% power to detect a population OR of 1.94 when comparing two groups of equal size.

In the FHSS, information on demographic characteristics, exposures and outcome variables was obtained simultaneously from all individuals within a narrow time period of 9 weeks. Some exposures such as age at first intercourse, condom use at sexual debut, and lifetime number of sexual partners reflect earlier exposures. In a cross-sectional study, only associations between variables can be assessed, and temporality or causality cannot be inferred [99]. Previous chlamydia test results could have influenced later sexual behaviour in the direction of less or increased risk causing a slight attenuation in the observed odds ratio estimates.

Infectious disease epidemiology has some unique features: a case may also be a risk factor, and a case may be a source without being recognised as a case due to asymptomatic infection [119]. Unprotected sexual intercourse with an infected subject is required for the occurrence of effect; a positive test result for *C. trachomatis*. As an infected subject is a source of disease in others, contact patterns in society, i.e. who meets whom, how do they meet, and how often, are important issues in order to understand the chlamydia epidemiology in a population [119].

5.3.1 C. trachomatis genotyping

Paper I describes the application of a high-resolution typing method in 248 *C. trachomatis* samples; 60 samples from the adolescent general population in Finnmark and 188 samples from the clinical laboratory routine in Trondheim, Tromsø and Finnmark (Figure 4). We detected a striking genetic diversity; 50 different STs with more than half observed in single individuals only, while two-thirds were found in less than four individuals. Multiple STs and alleles were novel. This is in line with a recent study using this MLST scheme on chlamydia samples from 52 heterosexual STI clinic patients in an previously unmapped area [114], and has also commonly been observed in databases for other bacteria [116]. The minimum spanning tree analysis showed that most STs belonged to clonal complexes which corresponds to other bacterial MLST databases [116]. MLST displayed a 4.5 higher resolution than *ompA* sequencing revealing a larger genetic diversity.

The use of Simpson's diversity index, *D,* provides a method to calculate the probability that two strains sampled at random from the test population will be placed into different typing groups and has most value for large and non-local collections of strains [93]. As the 60 high school samples were collected from adolescents in 5 schools during 9 weeks and the high prevalence of specific STs indicated presence of shared sexual networks, it was likely that some transmission of *C. trachomatis* infections between study participants had occurred, thus violating the criteria of epidemiological independency. We therefore calculated Simpson's *D* only in the 188 routine clinical samples. There may also have been a dependency structure in these samples due to the sampling context in the laboratories (short time period, only age group 15-20 years, defined geographic areas) which could lead to an underestimation of the diversity index *D.* Accordingly, both *D* and an adjusted D_c were calculated.

Only 1.6% of specimens were identified as nvCT that caused a clonal outbreak in Sweden in 2007 [22, 40]. The low prevalence was as expected because nvCT has rarely been found

outside Sweden [22, 117, 118]. After appropriate laboratory testing was introduced in Sweden, a selective decline of 24-40% of nvCT occurred in several counties suggesting a further decreased potential for nvCT to spread across international borders [120, 121].

Genovar E comprises about half of genital chlamydia infections in heterosexual women and men, a distribution which has appeared stable over time and geography [24-28]. Genovar G has been associated with infection transmission in networks of men who have sex with men [115, 122]. The FHSS was the first to report an atypical genovar distribution with G comprising 36% in a mainly heterosexual adolescent population. This was mostly due to the high occurrence of ST128 and the unique ST161. We assumed that ST128 and ST161 had expanded in local sexual networks in Finnmark, but lack of sexual network data hampered further investigation. ST161 has not been identified in chlamydia samples from other catchment areas analysed in Uppsala, Sweden, after our study in 2009 (Björn Herrmann, personal communication).

The *ompA* gene is shown to undergo extensive recombination [1, 24, 123], but the frequency of recombination events is yet unknown (Nicholas R. Thomson, the Sanger Institute, UK, personal communication). Recent whole-genome sequencing (WGS) studies by Harris *et al.* found that *ompA* has been transferred between phylogenetically unrelated *C. trachomatis* strains on several occasions providing evidence of its unsuitability as a marker for strain typing [1, 19, 124].

We found ST12, ST128 and ST161 to be the most frequent and thus most successful genotypes in Finnmark. Their success could be explained by a high transmission rate or by these STs causing a silent infection. However, only half of these infections were asymptomatic. As only 60 samples carried behavioural data, the reasons for the success could not be further elucidated. ST distribution in sexually active adolescent populations needs to be

studied in larger populations with more chlamydia samples and inclusion of partner characteristics and sexual network data.

We concluded that the MLST scheme is a valuable tool for studying the molecular epidemiology of *C. trachomatis* infections and far superior to *ompA* sequencing in terms of resolution. The MLST method has later been modified by the use of shorter target regions and nested PCR for increased sensitivity [114]. Stability and reproducibility of the 5 targets after multiple passages of nvCT in cell culture have also been documented [125]. The method is still too labour intensive to be used for partner notification in clinical practice, but next generations' sequencing technology may increase speed and make it more affordable.

All MLST schemes are limited by the fact that they only use a small fraction of the genome, and samples that are indistinguishable with respect to the 5 target regions may have considerable variation in the remaining DNA [94]. MLST only occasionally detects mixed chlamydia infections. The ultimate chlamydia typing method would be WGS with robust and finished sequences that are generated economically and quickly. A recent study showed that it is now possible to obtain WGS directly from clinical samples without culture [123]. As WGS is becoming cheaper, faster, high throughput and more available, it may in the future be one of the preferred genotyping methods and be performed in samples from clinical practice [19]. However, getting enough DNA for WGS will always be challenging, and future genotyping using WGS may be developed in two phases. First, gathering a large number of samples with as high diversity as possible to obtain WGS data. Second, from WGS data develop an SNP (single nucleotide polymorphism) based typing scheme with suitable resolution for molecular epidemiology. The SNP-scheme will be preferable to WGS if there is limited DNA (N. R. Thomson, personal communication).

5.3.2 Early sexual behaviour and chlamydia infection

In Paper II, we detected significant differences in sexual behaviour between genders. Chlamydia prevalence among sexually active girls was 7.3% and among boys 3.9% in agreement with the high IRs in surveillance data from Finnmark. Corresponding gender ratios were observed in the high school based screenings in Philadelphia (girls 8.1% and boys 2.5%) [49], in New York (8.9% and 3.8%) [82], and in the first round of screening in New Orleans (11.5% and 6.2%) [81]. A similar gender ratio, but significantly lower prevalence was observed in the school based screenings in San Francisco (2.2% and 0.6%) [44], Luxembourg (2.5% and 0.9%) [48], and in a low-incidence area in Southern Norway (2.9% and 1.0%) [47] indicating that high school based screening may not be efficient use of resources in low-morbidity areas.

In our study, we confirmed that girls started to have sex at an earlier age than boys, more often were in steady relationships, had older partners, reported less time since last intercourse (results not shown) and were poorer condom users than boys at this occasion, which is in line with other Nordic studies on adolescent sexual behaviour [62-67, 126]. More boys than girls reported casual last sexual partners (21% vs 11%, $p < 0.001$) and more alcohol use overall and related to last sexual intercourse, also in agreement with other studies [62, 63, 67, 126]. Accordingly, girls and boys had different risk profiles for infection.

Adolescent males reporting higher number of sexual partners than females is what particularly has separated genders in their responses in sexual behaviour surveys [62, 63, 81, 109]. In contrast, girls and boys in the FHSS reported similar number of sexual partners past 6 months. As girls had earlier sexual debut, more boys than girls reported 1-2 lifetime partners, while more girls than boys reported $\geq$ 6 lifetime partners. The results may indicate a new cohort of more sexually active and self-confident adolescent females as also suggested by others [67]. In contrast, recent studies in the same age group in Southern Europe found that adolescent

boys still report earlier sexual debut and higher lifetime number of partners than girls [55, 127, 128].

Among girls, several previously well-documented risk factors increased the odds of prevalent chlamydia infection in crude and multivariable analyses: ethnicity (Sami/Sami-Norwegian), ≥2 sexual partners past 6 months, and ≥6 lifetime partners [51, 112, 129]. In addition, higher maternal education and meeting venue for last partner increased odds of infection. We found that daughters of higher educated mothers reported more substance use overall and in connection with last intercourse than those with less educated mothers, which may suggest that higher educated women leave their daughters more freedom. The increased infection risk associated with particular meeting venues could reflect high-risk sexual behaviours and increased prevalence among persons frequenting these venues [130].

In boys, no condom use at first intercourse and having had an older last sexual partner increased the odds of infection in crude and multivariable analyses. Condom use at sexual debut was a strong predictor of condom use at last intercourse. Only 12% of the boys reported older last sexual partner, but this increased the odds of infection threefold in boys, which is similar to results from a recent study in the US [131]. This odds ratio became non-significant when adjusting for lifetime number of partners, indicating that adolescent boys who attract older women are more sexually active than peers with same-aged or younger partners.

In the multivariable model for girls and boys combined, residence outside the family home during school year, ≥2 partners past 6 months, meeting last partner at a party, bar or disco, and no condom use at last intercourse increased the odds of prevalent infection. One third of participants had left their home municipality to attend high school and they had twice the odds of infection compared to students living at home. The reasons may be less parental and societal control. A 1979 school survey on adolescent sexual behaviour and contraceptive use

in a Finnmark municipality found that girls living outside their family home were more sexually active than those living at home [66]. We found no other studies that have examined associations between residence during high school and prevalent chlamydia infection.

Due to detection of significantly more gender-specific genotypes (STs) in girls than boys, and most girls reporting older last sexual partners, we concluded that a majority of girls were linked to off-school sexual networks with assumed higher chlamydia rates as indicated by surveillance data (Paper I and II). Unlike what we expected, sexual partner age was not significantly associated with female chlamydia infection. The variable 'partner age' was based on age of participants' last sexual partner. Girls reporting same-aged or younger sexual partners may have had recent older last partners, thus obscuring the association between partner age and infection risk in girls.

In Paper II, we showed that accumulation of gender-specific early sexual experiences may contribute to a different chlamydia risk profile in girls and boys. Nagelkerke's estimate of explained variance in the multivariable models with outcome variable chlamydia infection was 13% in girls, 19% in boys, and 14% in both genders combined. We may have missed important behavioural chlamydia risk factors like concurrent sexual partnerships not assessed in the questionnaire [111-113]. We concluded that the genders are vulnerable to infections at different times during adolescence due to differing behaviour with girls on average initiating their sexual careers and being more sexually active at a younger age than boys. This suggests the need for gender-specific interventions in this age group.

5.3.4 Chlamydia testing in a high school based screening and previously in clinical practice

In Paper III, we detected significant gender differences in previous clinic based testing as more than half of sexually active girls had been tested compared to only one-fifth of boys. This is in agreement with national surveillance data and previous studies in Norway and other

high-income countries having implemented extensive chlamydia testing or screening programmes [38, 43, 51, 79, 132].

The school based screening reached a high and similar proportion of both genders and proved efficient in terms of proportion of population tested, and number of infections detected and treated. The following factors were assumed to be important for the unusually high participation in the school based screening: thorough planning, the acceptance gained from the principals, the information provided to teachers, students and parents before data collection, the relevant topics, the universal offer to all students irrespective of sexual history, the 'in-class' recruitment and sampling procedures, the efficient logistics, rapid notification of positive test results, and FHSS being the first chlamydia high school based screening in Northern Norway [98, 133]. It is likely that invitation to participate in research increased uptake. External researcher led recruitment may also have contributed [134].

A high proportion of boys accepting the offer to be tested has also been observed in similar studies [47, 49, 82, 135]. Previous studies in non-clinical settings have found that easy access, convenient testing procedures, high levels of confidentiality and individual provider characteristics may influence boys' decision to be tested [78, 80, 136-139].

Female participants in the FHSS had high chlamydia prevalence irrespective of previous clinic based testing; 7.3% if previous clinic based testing versus 7.2% if school-only test. Girls with clinic based testing had higher levels and longer duration of sexual risk behaviours than those with school-only test (p-values <0.03 for the variables; $\geq$2 sexually active years, $\geq$2 partners past 6 months, $\geq$6 lifetime partners, older last partner, and no condom at last intercourse). The equal chlamydia prevalence may indicate effect of adherence to testing and treatment recommendations in girls with clinic based testing. This assumption is supported by the natural experiment that occurred in Sweden with nvCT that escaped detection and was

able to spread freely in mostly younger age groups [40, 120, 121]. Diagnosis, treatment and partner notification was discontinued in nvCT cases since they tested false negative. After correction of the diagnostic targets, a rebound chlamydia epidemic was observed (Figure 1) strongly indicating that early diagnosis and treatment does affect community transmission and decrease prevalence [120]. Alternatively, the two groups of girls had equal prevalence due to unrecognised risk factors in the school-only test group.

Boys with previous test had about the same prevalence as girls. The finding that boys with school-only test had less than half the prevalence of girls is consistent with less sexual activity in boys this age and hence reduced risk of infection (Paper II). Half of chlamydia infections were detected in the school-only test group, and correspondingly participants testing positive reported higher levels of sexual risk behaviours than participants with school-only test and negative test results.

High school based screening and previous clinic based testing were associated with completely different independent variables. In the multivariable analysis of girls and boys combined, known chlamydia risk factors such as female gender, young age at sexual debut, no condom use at first intercourse, and higher number of lifetime sexual partners increased the odds of clinic based testing. This indicates that these adolescents were aware of behavioural STI determinants. Among boys, testing varied by ethnic group which is in line with a recent Dutch study [79]. Nagelkerke's estimate of explained variance in the multivariable model for all participants with outcome 'clinic based testing' was 42% showing a good model fit, indicating that important variables were included in the model.

In contrast, several factors unknown to increase chlamydia infection risk in adolescents such as male gender, academic affiliation, later sexual debut, and condom use at first intercourse increased the odds of school based screening in the multivariable analysis of both genders

combined. The school based screening participants accepted test services they were not seeking. Based on the New Orleans school screening, Nsuami *et al.* have suggested that persons accepting school based STI screening are motivated by a collective acceptance of something that is being offered to everybody, rather than by a rationalisation of individual chlamydia risk [140].

Among all participants, 42% of girls and 7% of boys reported current urogenital symptoms, and this variable was observed to have the strongest association with school based screening in the multivariable analysis of both genders combined. However, 92% of participants without symptoms and 97% with symptoms were tested indicating high participation among those without urogenital complaints. A Nagelkerke's estimate of 6.2% may also reflect that accepting school based screening is not motivated by participants' sexual risk profile. Boys tested in the FHSS frequently commented on the convenience of 'everybody getting tested', which also has been emphasised by male participants in other non-clinical screening studies [80]. Engaging boys in chlamydia testing early on may constitute a preventive strategy for their female partners, normalise testing, and increase young men's interest in ensuring their own sexual health [111].

High school students represent easily accessible populations. Repeat annual screening and treatment with high enough participation should theoretically have the potential to reduce the transmission and reservoir of chlamydia infections in the target population [3, 141]. However, participation in the repeat high school based screening programme in New Orleans declined from an initial 56% in 1995-96 down to 33% in the final year 2004-05 [135]. This was mainly due to decrease in number of students with parental consent. Only a limited number of students were enrolled for multiple years and sexual behaviour was recorded only for a few years. In New Orleans, repeat screening was not associated with significant change in

chlamydia prevalence in females or males among those who were tested more than once [142]. In Philadelphia, there was only a slight decline in female positivity from 8.4% in the first year to 7.2% in year 5, while male positivity remained at 2.5% [143]. The failure to reduce prevalence has been explained by incomplete coverage, links to off-school sexual networks with higher infection prevalence, inability to reach high-risk core group members, inadequate partner notification and treatment, and insufficient screening frequency [141].

Repeat high school based chlamydia screening has not been tried in Norway, but repeat high school studies on adolescent health and lifestyle including biological samples have shown sustained response rates above 85% [133, 144]. In addition, parental consent only being required in participants <16 years, most students attending three consecutive school years, the homogeneous nature of the Norwegian society, the liberated attitudes towards adolescent sexuality, and the high participation rate in the FHSS may suggest a potential for sustained higher participation in repeat school based screenings in Norway compared to the US [135].

In the past years, the evidence that asymptomatic lower genital chlamydia infection is likely to cause pelvic inflammatory disease and reproductive complications has been questioned [11, 145-147] and consequently the evidence that supports large screening programmes in the population [73, 148-150]. Given the high participation rate in the FHSS that provided access to almost entire birth cohorts of adolescents, repeat high school based studies on changing trends in sexual behaviour using chlamydia infection as a biomarker for risk behaviour may still be valuable. The results could be used to develop innovative targeted interventions to increase safe sexual behaviour among adolescents. We thus suggest conducting repeat high school based screenings in selected high-morbidity areas designed as research studies with continuous evaluation of feasibility, cost, participation, and effect on prevalence [151].

6. Conclusions

In summary, our findings suggest that:

- MLST of *C. trachomatis* had significantly higher resolution than traditional *ompA* genotyping and enabled the detection of specific STs such as the Swedish nvCT. We found multiple novel alleles, new STs, and unique STs in line with studies on different bacteria in previously unmapped geographic areas and in accordance with other newly established bacterial MLST databases. There were no significant differences in genetic diversity of STs between the three areas. Due to high resolution and detection of specific STs, we concluded that MLST is a useful tool in molecular chlamydia epidemiology.

- The high chlamydia prevalence of 5.7% detected in the FHSS corresponded to the high annual IR in surveillance data observed in Finnmark. Gender differences in sexual behaviour in the early sexually active years contributed to gender differences in risk profiles for chlamydia infection. This probably contributed to girls having twice the prevalence of boys. Girls and boys being vulnerable to chlamydia infections at different times during adolescence due to behavioural factors suggests the need for gender-specific chlamydia control strategies in this age group.

- Threefold more adolescent girls than boys reported previous clinic based chlamydia testing. Previous testing was associated with mostly known chlamydia risk factors suggesting awareness of behavioural determinants. An unusually high and equal proportion (93%) of sexually active girls and boys were tested in the school based screening, which was mostly associated with factors unknown to increase infection risk. Girls had high and equal prevalence independent of previous testing. Half of chlamydia infections were detected in participants never tested. The high participation

and detection and treatment of a large chlamydia reservoir suggests school based screening as a potential tool to decrease chlamydia transmission among sexually active adolescents in high-morbidity areas in Norway.

7. Implications for future research

The findings in this thesis related to genotyping, participation, chlamydia prevalence, sexual behaviour, and testing patterns suggest several interesting questions to be further explored.

The FHSS sexual behaviour data (sexual debut age, circumstances related to first intercourse, attitudes and feelings towards sex among those without sexual debut) will be further explored in separate papers, and so will the associations between chlamydia infections and self-assessed risk, condom use, and sexual behaviour on the Internet.

When possible, WGS should be applied to chlamydia samples collected in a general population. WGS data should be linked to questionnaire data on urogenital symptoms, sexual behaviour, and sexual networks. The unique ST distribution in Finnmark should be further examined in a repeat study including a larger study population and more chlamydia samples. WGS should in particular be applied to ST161/genovar G chlamydia samples.

In future studies, self-collected vaginal swabs rather than FVUs should be used to increase sensitivity in the female samples. The sexual behaviour data should be more comprehensive and include different sexual practices (vaginal, oral and anal intercourse), concurrent relationships, and sexual partner characteristics such as school enrolment, ethnicity, country of origin, and risk assessment of partner. Sexual partners should be included in the study for prevalence detection and genotyping of positive specimens. Previous clinic based testing should be validated using laboratory data. Partner treatment rates should be assessed. High schools in other high-morbidity areas in Norway should be included to increase generalisability. Repeat high school based screening studies including comprehensive sexual behaviour questionnaires in selected high-morbidity areas should be tried and evaluated.

8. References

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Paper I

Paper II

Paper III

Appendix A

Information letter

English translation

KORSN
KOMPETANSESENTER I SMITTEVERN HELSE NORD RHF

Invitation to participate in a research project in fall 2009

'Sexual behaviour and chlamydia among high school students in Finnmark'

Background and purpose

This is an invitation to participate in a research study on sexual behaviour and chlamydia prevalence among high school students in Alta, Hammerfest, Sør-Varanger, Karasjok and Kautokeino. The study is conducted by the Competence Centre of Infection Control in the Regional Health Authority of North Norway, University Hospital of North Norway, and by the University of Tromsø.

Chlamydia is a common sexually transmitted infection (STI) among Norwegian adolescents. Over the years, Finnmark and Troms counties have reported higher chlamydia rates than the rest of Norway. Chlamydia infection often is asymptomatic and you can be infected without recognizing it. Untreated infection can lead to infertility, ectopic pregnancy, and chronic pelvic pain in females.

The study results will provide important knowledge on the high chlamydia rates among adolescents in Finnmark and the public health measures necessary to control infection rates.

What does study participation imply?

The participants are asked to fill in a web-based questionnaire during class hours in the presence of a teacher and two persons from the research staff. The questionnaire includes questions on future educational plans, lifestyle, attitudes towards sex, contraceptive use, and much more. It is important to answer all questions as honestly as you can. Several questions focus on personal matters. You do not have to complete this questionnaire or any question if you do not want to. It should take you about 20 minutes to fill in all answers. The questionnaire is completely confidential so nobody will see the answers you give. You will not fill in your name or personal identity number in the questionnaire.

Directly after finishing the questionnaire, you continue to the school toilet and provide a urine sample. Sampling equipment will be handed out in class. We ask all participants to provide a sample, including those who have not yet had their sexual debut.

The urine sample will be analysed at the University Hospital of North Norway in Tromsø. Participants with a positive chlamydia test result will receive a text message on their mobile phone. If we receive no reply from you, we will call you on your mobile phone. Eventually, we will send a letter by mail asking you to call us. *We will not send a letter with a positive test result to your parents' house.*

The local health care service will provide a prescription on antibiotics free of charge to participants with a positive chlamydia test result. Sexual partners will be contacted for testing and treatment.

If approved by the Data Protection Authority and the National Regional Committee for Medical and Health Research Ethics, data on participants testing positive for chlamydia will be linked to the Norwegian Prescription Database to examine if participants did fill in the prescription for antibiotic. These data will not include personal identifiable information when received by the researchers.

All participants will be included in a lottery with 3 persons winning an exclusive mobile phone.

Advantages and disadvantages

You will have a free chlamydia test. If you test positive, you will receive treatment. The information in the questionnaire will provide important knowledge on chlamydia infections among adolescents in Finnmark and the public health measures needed to reduce the epidemic.

You may find some of the questions too sensitive to answer. You do not have to answer all questions.

What will happen to the urine sample and the personal information?

The data will be used for research purposes only. All the analysed data will be person non-identifiable and will not be linked to your name or your personal identity number.

We use QuestBack online survey system. The data will be safely stored on servers at the University Hospital of North Norway (UNN) in Tromsø and can only be accessed by the project manager.

Urine samples testing positive for chlamydia will be analysed using a 'finger print' technique to map the chlamydia clones present in adolescents in Finnmark and compare to international clones.

Voluntary participation and consent

We must stress that your participation is voluntary. Students 16 years or older should consent to participation by filling in the questionnaire. Students younger than 16 years have to provide written consent signed by their parents or guardians. You may withdraw from the study at any time and without stating any reason.

Students choosing not to participate can do school work while the others are filling in the questionnaire. School attendance is mandatory while the study is conducted. Absence will be registered by the teacher.

Please, contact us if you choose to withdraw from the study or if you have any questions: Project manager Kirsten Gravningen, phone: 776 27044/ email: kirsten.gravningen.@unn.no or public health nurse/ research assistant Randi E. Olsen, phone: 776 69552/ randi.elisabeth.olsen@unn.no.

Privacy policy and security

All information given in the questionnaire is strictly confidential. The staff is bound by confidentiality. UNN Tromsø by the Managing Director is responsible for data management. The data management and security system is approved by the Data Protection Official at UNN.

Bio-bank

Urine samples with a negative chlamydia test result are thrown after two days. Samples with a positive test result will be stored until the 'finger print' analysis is complete and then be thrown away. If you consent to participate in the study, you also consent to temporal storage of a chlamydia positive urine sample until January 2010.

The right to access and delete personal data and the deletion of samples

If you consent to participation, you have the right to view your personal information, and errors can be corrected. If you withdraw from the study, you can request deletion of the collected data unless the data already are included in analyses or in scientific publications.

Economy

The study is funded by Sparebank1 Nord-Norges Medical Research Grant, The Norwegian Directorate of Health, and The North Norway Regional Health Authority.

Information about the study results

The study results will be published at research conferences/ meetings and in scientific articles based on larger groups, and will not be stratified by class or school.

Ethics

The study is approved by the National Regional Committee for Medical and Health Research Ethics.

Duty of confidentiality

If you are younger than 16 years and have a positive chlamydia test result, we will contact only you for treatment and your parents/guardians will not be informed. The information in the questionnaire is subject to duty of confidentiality towards the parents according to The Norwegian Health Research Act §17-4.

Consent to study participation

Students younger than 16 years, have to provide written consent from the parents or guardians to participate in the research study.

I hereby consent to my daughter/ son ……..…… (*name)* participating in the study.

……..

(Signed by parents/guardians, date)

Appendix B

Information letter

Original Norwegian version

Forespørsel om deltakelse i forskningsprosjektet høsten 2009
"Seksualitet og klamydia blant elever i videregående skole i Finnmark"

Bakgrunn og hensikt
Dette er forespørsel om å delta i en studie som skal undersøke seksualvaner og forekomst av klamydia blant elever i videregående skole i Alta, Hammerfest, Sør-Varanger, Karasjok og Kautokeino. Studien utgår fra Kompetansesenter i smittevern Helse Nord, Universitetssykehuset i Nord-Norge og Universitetet i Tromsø.

Klamydia er en seksuelt overført infeksjon (kjønnssykdom) som er svært vanlig blant norsk ungdom. Finnmark og Troms har gjennom årene rapportert høyere forekomst av klamydia enn resten av landet. Ofte gir klamydia få plager, og det gjør at man kan være smittet med klamydia uten å vite det. Ubehandlet klamydiainfeksjon kan seinere i livet gi kvinner komplikasjoner som barnløshet, svangerskap utenfor livmoren og smerter i bekkenet.

Resultatene fra denne studien vil gi viktig informasjon om bakgrunnen for den høye forekomsten av klamydia blant ungdom i Finnmark, og hvordan helsetjenesten kan styrke det forebyggende arbeidet mot klamydia. Studien gjennomføres i skolene høsten 2009.

Hva innebærer studien?
Deltakerne besvarer et web-basert spørreskjema i klasserommet i skoletida. Læreren og to personer fra prosjektet vil være til stede. Spørreundersøkelsen dreier seg om fremtidsplaner, livsstil, holdninger til sex, prevensjonsbruk, med mer. Det er viktig at man besvarer spørsmålene så ærlig som mulig. Flere spørsmål handler om personlige forhold. Er det spørsmål man ikke kan eller vil svare på, lar man være. Det tar maks. 20 minutter å besvare skjemaet. Alle svarene er konfidensielle. Ingen lærere eller andre med tilknytning til skolen får se skjemaet etter at det er fylt ut. Man skal ikke fylle ut navn eller fødselsnummer på skjemaet.

Rett etter besvaring av skjema, går man til skolens toalett og avgir en urinprøve. Prøvetakingsutstyr deles ut i klassen. Av forskningsmessige hensyn er det viktig at alle deltakerne leverer urinprøve, også elever som ikke har debutert seksuelt.

Urinprøvene undersøkes for klamydia ved Universitetssykehuset i Tromsø. Funn av klamydia varsles til deltakeren *via sms på mobil*. Dersom vi ikke får kontakt via sms, vil vi ringe på mobilen. Siste utvei er å sende brev i nøytral konvolutt hvor vi ber om å bli kontaktet på et oppgitt telefonnummer. *Brev med positiv klamydiaprøve vil ikke sendes hjem til foreldrene.*

Lokal helsetjeneste vil sørge for behandling av klamydia ved å skrive resept på én-gangsdose med antibiotika. Behandlingen er gratis. Det vil gjøres smitteoppsporing blant partnere for å sikre at partnerne får tilbud om behandling.

Med forbehold om godkjenning fra Datatilsynet og Regional komité for medisinsk og helsefaglig forskningsetikk (REK nord), vil data om deltakere som tester positivt for klamydia, bli koblet mot opplysninger i Nasjonalt reseptregister for å undersøke om de har innløst resept på antibiotika fra apoteket. Data om deltakerne som har hentet ut antibiotika, vil utleveres til forsker uten person-identifiserende kjennetegn, bare som opplysninger om kjønn, alder og utleveringsdato for antibiotika.

Deltakerne i studien er med i loddtrekning om 3 fine mobiltelefoner i desember 2009.

Mulige fordeler og ulemper
En fordel med å delta er at du vil bli testet for klamydia og at du vil få nødvendig behandling hvis prøven er positiv. Opplysningene på spørreskjemaet vil bidra til å øke kunnskapen om den høye forekomsten av klamydia blant ungdom i Finnmark, og hvilke tiltak som kan settes inn for å forebygge og hindre klamydiaepidemien.

Undersøkelsen inneholder enkelte spørsmål som for noen deltakere kan oppleves som sensitive. Du trenger ikke å besvare alle spørsmålene i skjemaet.

Hva skjer med prøvene og informasjonen om deg?
Informasjonen som registreres, skal kun brukes slik som beskrevet i hensikten med studien. All informasjon vil bli behandlet uten navn, fødselsnummer eller direkte gjenkjennende opplysninger.

Prosjektet benytter Questback i gjennomføringen av spørreundersøkelsen. Svarene lagres på sikre servere Universitetssykehuset Nord-Norge (UNN) i Tromsø hvor kun prosjektleder har tilgang.

Urinprøver som er positive for klamydia, vil bli undersøkt nærmere med "fingeravtrykksanalyse"for å kartlegge hvilke kloner av klamydia som fins blant ungdom i Finnmark, og om det er de samme klonene som ellers i Norden.

Frivillig deltakelse og samtykke
Deltakelse i studien er frivillig. Elever som er fylt 16 år, vil gjennom å besvare spørreskjemaet gi sitt samtykke til å delta. Elever som *ikke har fylt 16 år*, må levere skriftlige samtykke fra foreldrene. Man kan når som helst og uten å oppgi grunn, trekke seg fra studien uten at dette får noen konsekvenser.

Elever som ikke deltar i studien, kan gjøre skolearbeid i klasserommet mens de andre besvarer spørreskjemaet. Det er *obligatorisk frammøte* til timen, og det føres fravær.

Dersom du senere ønsker å trekke deg eller har spørsmål til studien, kan du kontakte:
Prosjektleder Kirsten Gravningen, tlf dagtid: 776 27044/e-post til: kirsten.gravningen@unn.no.
Helsesøster/forskningsass. Randi E. Olsen, tlf dagtid 776 69552/ randi.elisabeth.olsen@unn.no.

Personvern og sikkerhet
Alle opplysninger som gis i spørreskjemaet, er konfidensielle. Ansatte i prosjektet har taushetsplikt. Universitetssykehuset Nord-Norge Tromsø ved administrerende direktør er databehandlingsansvarlig.

Datahåndtering og datasikkerhet er godkjent av Personvernsombudet ved UNN HF.

Biobank
Urinprøver som er negative for klamydia, kastes etter to dager. Urinprøver som er positive for klamydia, vil bli lagret inntil "fingeravtrykksanalyse" er utført. Deretter kastes også disse urinprøvene.

Hvis man sier ja til å delta i studien, gir man også samtykke til at urinprøven (hvis den er klamydiapositiv) og analyseresultatet oppbevares midlertidig til januar 2010.

Rett til innsyn og sletting av opplysninger om deg og sletting av prøver
Hvis man sier ja til deltakelse, har man rett til innsyn i egne registrerte opplysninger, og eventuelle feil kan korrigeres. Dersom man trekker seg fra studien, kan man kreve å få slettet innsamlet informasjon, med mindre denne allerede er inngått i analyser eller brukt i vitenskapelige publikasjoner.

Økonomi
Studien og biobanken er finansiert gjennom forskningsmidler fra Sparebank1 Nord-Norges Medisinske forskningspris 2008, Helsedirektoratet og fra Kompetansesenter i smittevern Helse Nord.

Informasjon om utfallet av studien
Resultater fra studien vil kunne publiseres som gruppedata på faglige konferanser/ møter og i vitenskapelige artikler uten at den enkelte klasse eller skole kan gjenkjennes. Resultatene vil brukes som grunnlag for å styrke tiltak mot seksuelt overførte infeksjoner blant ungdom i Finnmark.

Etisk godkjenning
Prosjektet er godkjent av REK nord.

Taushetsplikt
Dersom deltakere under 16 år får påvist klamydia, vil han/hun kontaktes direkte for behandling. Foreldrene til deltakere under 16 år vil *ikke informeres* om prøveresultatet. Prosjektet har også taushetsplikt overfor foreldre/ foresatte vedrørende opplysninger som eleven avgir i spørreskjemaet. Ovenstående er i henhold til helseforskningsloven § 17, 4. ledd.

Samtykke til deltakelse i studien

Elever som enda ikke er fylt 16 år, må levere skriftlig samtykke fra foreldrene til å delta i studien.

Jeg samtykker til at mitt barn ……..(*navn)* deltar i studien

(Signert av foreldre/foresatte, dato)

NB: Elever <u>under 16</u> år må ta utskrift av dette dokumentet og levere underskrevet samtykke til prosjektleder samme dag som studien gjennomføres i klassen.

Appendix C
Information letter
Original Sami version

Jearaldat searvat dutkanprošektii 2009 čavčča
"Seksualitehta ja klamydia Finnmárkku joatkkaskuvlaohppiid gaskka"

Duogáš ja ulbmil
Dá lea jearaldat searvat iskkadeapmái mas mii galgat iskkadit Álttá, Hammerfeastta, Mátta-Várjjaga, Kárášjoga ja Guovdageainnu joatkkaskuvlaohppiid seksuála dábiid ja lea go sis klamydia. Iskkadeami čađahit *Kompetansesenter i smittevern Helse Nord* (Davvi Dearvvašvuođa njoammuneastadeami Gelbbolašvuođaguovddáš), Davvi-Norgga Universitehtabuohcciviessu ja Romssa Universitehta.

Klamydia lea seksuálalaččat njommon infekšuvdna (nuoskkesdávda), mii lea ollu nuorain Norggas. Finnmárkkus ja Romssas lea dát dávda maŋemus jagiid gávdnon sakka eanet go muđui riikkas. Dávjá olmmoš ii dovdda ahte sus lea klamydia, ja dat dahká ahte olbmos sáhttá leat dávda vaikko ieš ii dieđe ge. Jus ii dálkkot klamydia, de sáhttá dat dagahit nissonolbmui maŋŋelis eallimis ahte ii sáhte oažžut máná, dahje ahte ohki (mánná) šaddagoahtá olggobeallái mánnágoađi, ja sáhttá maid dagahit bákčasiid vuolil.

Dán iskkadeami bohtosat addet deatalaš dieđuid dasa manne nu ollu Finnmárkku nuorain lea klamydia, ja movt dearvvašvuođabálvalus sáhttá hehttet klamydia leavvamis. Iskkadeapmi čađahuvvo skuvllain 2009 čavčča.

Máid sisttisdoallá iskkadeapmi?
Oasseváldit vástidit web-vuođustuvvon jearahallanskovi luohkkálanjas skuvlaáiggis. Oahpaheaddji ja guokte olbmo prošeavttas leat doppe dalle. Jearahallanskovis leat jearaldagat boahtteáiggi plánaid birra, eallinvuogi ja dan birra makkár oainnut dus leat sexii, prevenšuvnnaid geavaheapmái jna. Lea deatalaš vástidit gažaldagaid nu rehálaččat go vejolaš. Máŋga gažaldaga leat persovnnalaš dilálašvuođaid birra. Jus leat gažaldagat maid it hálit vástidit, de lea dat ortnegis. Ádjána eanemusat 20 minuhta vástidit jearaldagaid. Buot vástádusat dollojuvvojit čiegusin. Ii oktage oahpaheaddji, eai ge earát geat gullet skuvlii, beasa oaidnit vástádusaid. Skovvái ii galgga čállit nama, ii ge riegádanbeaivvi.

Dakkaviđe go leat vástidan skovi, galggat mannat skuvlla hivssegii ja addit gožžaiskosa. Mii addit luohkkálanjas doasa masa goččat. Dutkama dihte lea dehálaš ahte buot oasseváldit addet gožžaiskosa, maiddái sii geain ii leat vuos leamaš sexa.

Romssa Universitehta iská gožžaiskosiin lea go klamydia. Jus gávnnahuvvo klamydia, de mii dieđihit *sms bokte mobiltelefovdnii*. Jus eat oačča oktavuođa sms bokte, de riŋget mobiltelefovdnii. Maŋemus vejolašvuohta lea sáddet reivve nøytrála konvoluhtas, mas mii bivdit sáddet telefonnummára. *Reive mii dieđiha ahte dus lea klamydia, ii sáddejuvvo ruoktot du váhnemiidda.*
Báikkálaš dearvvašvuođabálvalus addá divššu klamydia vuostá dan bokte ahte čállá resepta antibiotika-dálkasii maid galggat okte váldit. Dikšu lea nuvttá. Mii fertet de diehtit geainna/geaiguin son, geas gávnnahuvvo klamydia, lea ovttastallan, vai beassat maiddái su/sin dikšut.

Jus Databearráigeahčču ja medisiinnalaš ja dearvvašvuođafágalaš dutkanetihkka (REK nord) dohkkehit, de datat oasseváldiid birra geain lea klamydia, čadnojuvvojit dieđuide Našuvnnalaš reseptaregistarii iskan dihte leat go sii viežžan antibiotika apotehkas. Diehtu geat vižžet antibiotika resepttain maid leat ožžon, sáddejuvvo dutkiide almmá nama haga ja eará dieđuid haga mat sáhtáše gávdnat geat sii leat. Dutkit ožžot dušše dieđuid agi ja sohkabeali birra ja guđe dáhtona lea viežžan antibiotika-dálkasa.

Sii geat servet iskkadeapmái leat mielde vuorbádeamis, mas geassit golbma fiinna mobiltelefovnna juovlamánus 2009.

Vejolaš ovdamunit ja heajut bealit
Okta ovdamunni lea ahte beasat iskkahit lea go dus klamydia ja ahte oaččut dárbbašlaš divššu jus dus lea. Dieđut maid mii oažžut jearahallanskovi bokte veahkehit min oahppat eambbo dan birra manne nu ollu Finnmárkku nuorain lea klamydia, ja movt sáhtášii hehttet klamydia leavvamis ja movt hehttet klamydia-epidemiija.

Iskkadeamis leat gažaldagat mat soitet soames ohppiid mielas leame menddo persovnnalaččat. Don it dárbbaš vástidit buot gažaldagaid, jus it hálit.

Mii geavvá iskosiiguin ja dieđuiguin du birra?
Dieđut maid registreret, galget adnot dušše nu movt lea čilgejuvvon iskkadeami ulbmilis. Dieđut eai sáhte čadnot nammii, riegádannummarii eai ge eará dieđuide mat sáhttet muitalit gii lea máid vástidan.

Prošeakta geavaha Questback jearahallaniskkadeami čađaheamis. Vástádusat vurkejuvvojit Davvi-Norgga Buohcciviesu (UNN) sihkkaris serváriidda, gosa dušše prošeaktajođiheaddji beassá.

Gožžaiskosat main gávdno klamydia, iskkaduvvojit dárkileappot nu gohčoduvvon "suorbmaluoddaanalysa" bokte, gávnnahan dihte makkár klamydia-klonat Finnmárkku nuorain leat, ja leat go dat seammaláganat go muđui Davviriikkain.

Eaktudáhtolaš searvan ja mieđiheapmi
Iskkadeapmái searvan lea eaktudáhtolaš. Oahppit geat leat deavdán 16 jagi, mieđihit searvamii dan bokte ahte vástidit gažaldagaid. Oahppit geat *eai leat deavdán 16 jagi*, fertejit addit váhnemiid vuolláičállaga. Sáhtát vaikko goas geassádit iskkadeamis, it ge dárbbaš muitalit manne, ii ge das leat mihkke váikkuhusaid dutnje.
Oahppit geat eai searvva iskkadeapmái sáhttet bargat skuvlabargguid luohkkálanjas dan botta go earát vástidit jearaldagaide. Lea *bákkolaš boahtit diibmui*, ja jávkan merkejuvvo.

Jus maŋŋil háliidat geassádit dahje jus leat gažaldagat iskkadeami birra, de sáhtát váldit oktavuođa:
Prošeaktajođiheaddji Kirsten Gravningen, tlf beaivet: 776 27044/e-poasta: kirsten.gravningen@unn.no.
Dearvvašvuođadivššár/dutkanassisteanta Randi E. Olsen, tlf beaivet 776 69552/ randi.elisabeth.olsen@unn.no.

Persovdnasuodjalus ja sihkarvuohta
Buot dieđut mat addojuvvojit jearahallanskovis dollojuvvojit čiegusin. Prošeavtta bargiin lea jávohisvuođageasku. Davvi-Norgga Buohcciviessu Romssas, hálddahusdirektora bokte, lea dat geas lea ovddasvástádus gieđahallat dieđuid.
Dieđuid gieđahallama ja datasihkarvuođa lea UNN HF Persovdnasuodjalusáittardeaddji dohkkehan.

Biobáŋku
Gožžaiskosat main ii leat klamydia, bálkestuvvojit 2 beaivvi maŋŋel. Gožžaiskosat main lea klamydia, vurkejuvvojit dassážii go "suorbmaluoddaanalysa" lea čađahuvvon. Dasto bálkestuvvojit maiddái dát gožžaiskosat.

Jus mieđihat searvat iskkadeapmái, de seammás dohkkehat ahte gožžaiskkus (jus das lea klamydia) ja analysaboađus vurkkoduvvo gitta ođđajagemánu 2010 rádjái.

Vuoigatvuohta diehtit ja sihkuhit dieđuid iežat birra ja iskosiid duššindahkat
Jus mieđihat searvat iskkadeapmái, de dus lea maid vuoigatvuohta oažžut diehtit makkár dieđut leat registrerejuvvon du birra ja vejolaš boasttuvuođaid sáhtát njulget. Jus geassádat iskkadeamis, de sáhtát gáibidit ahte čohkkejuvvon dieđut du birra sihkkojuvvojit, jus dat juo eai leačča oassin analysain dahje geavahuvvon dieđalaš čállosiin.

Ekonomiija
Iskkadeapmi ja biobáŋku leat ruhtaduvvon dutkanruđaid bokte maid Seastinbáŋku 1 Davvi-Norga lea juolludan Medisiinnalaš 2008 dutkanbálkášumi bokte, ja maiddái Dearvvašvuođadirektoráhtta ja *Kompetansesenter i smittevern Helse Nord* (Davvi Dearvvašvuođa njoammuneastadeami Gelbbolašvuođaguovddáš) leat ruđalačča dorjon prošeavtta.

Dieđut iskkadeami bohtosa birra
Iskkadeami bohtosiid sáhttá almmuhit oppalaš diehtun fágalaš konferánssain/čoahkkimiin ja dieđalaš artihkkaliin, nu ahte ii sáhte dovdat ovttaskas skuvlla dahje luohká. Bohtosiid sáhttá atnit nannet doaimmaid seksuála dávddaid njoammuma vuostá Finnmárkku nuoraid guovdu.

Ehtalaš dohkkeheapmi
REK nord lea dohkkehan prošeavtta.

Jávohisvuođa doallan
Jus vuollel 16 jagi oasseváldis gávnnahuvvo klamydia, de váldit suinna njuolga oktavuođa divššu birra. Vuollel 16 jahkásaš oasseváldiid váhnemiidda *ii dieđihuvvo* iskkadeami boađus. Váhnemat/fuolaheaddjit eai ge oačču diehtit maide eará das maid oahppi vástida jearahallanskovis. Dát lea dearvvašvuođadutkama lága § 17, 4. lađđasa mielde.

Miedđiheapmi searvat iskkadeapmái

Oahppit geat eai leat vuos deavdán 16 jagi, fertejit addit skovi, masa váhnemat leat miedđihan ahte searvvat iskkadeapmái.

Mun suovan iežan máná ……..(*namma)* searvat iskkadeapmái

(Váhnemiid/fuolaheddjiid vuolláičála, beaivi)

NB: Oahppit <u>vuollel 16</u> jagi fertejit prentet dán dokumeantta ja addit prošeaktajođiheaddjái vuolláičállojuvvon skovi seamma beaivvi go iskkadeapmi čađahuvvo skuvllas.

Appendix D
Questionnaire
English translation

Research study:
Sexual behaviour and chlamydia in Finnmark

First, we'd like to have some information about your background and how you look at yourself

---

1) * Gender

☐ Boy/man ☐ Girl/woman

2) * Year of birth, four digits (YYYY):

3) * School municipality

Select answer

4) Where do you live (residence) during the school year?

- ☐ At home with my parents/guardians
- ☐ Grandparents/other relatives
- ☐ Private room/appartment
- ☐ Student house
- ☐ Host family
- ☐ Other

Education

5) What is the highest level of education you plan to complete?

- ☐ High school (academic affiliation, sport, music-dance-drama)
- ☐ Vocational school
- ☐ College or university <=4 years (ex: bachelor, teacher, police, nurse, engineer, journalist)
- ☐ College or university >4 years (ex: master's degree, lawyer, civil engineer, doctor, dentist)
- ☐ I have not decided yet
- ☐ Other

6) What level of education did your mother complete?

- ☐ 9 (7) years of elementary school
- ☐ Vocational school
- ☐ High school degree
- ☐ College or university <=4 years
- ☐ College or university >4 years
- ☐ Don't know

7) What level of education did your father complete?

- ☐ 9 (7) years of elementary school
- ☐ Vocational school
- ☐ High school degree
- ☐ College or university <=4 years
- ☐ College or university >4 years
- ☐ Don't know

Culture and contact

8) My perceived ethnicity is: (Tick all options that apply)

☐ Norweg. ☐ Sami ☐ Russian ☐ Kven ☐ Finnish ☐ Other

9) Ethnicity of grandparents, mothers' side: (Tick all that apply)

☐ Norweg. ☐ Sami ☐ Russian ☐ Kven ☐ Finnish ☐ Other

10) Ethnicity of grandparents, father's side: (Tick all that apply)

☐ Norweg. ☐ Sami ☐ Russian ☐ Kven ☐ Finnish ☐ Other

11) Do you and your parents/guardians have a religious affiliation?

	Church of Norway	Laestadian	Jehovas witnesses/ Pentecostals	Russian orthodox church	Islam	No affiliation
Myself	☐	☐	☐	☐	☐	☐
Mother	☐	☐	☐	☐	☐	☐
Father	☐	☐	☐	☐	☐	☐

Self-Esteem

The following questions address whether you have negative thoughts about yourself. For each statement, please tick the most suitable category.

12) Thinking negatively about myself is something ...

	Strongly agree	Agree	Don't know	Disagree	Strongly disagree
I often do	☐	☐	☐	☐	☐
I authomatically do	☐	☐	☐	☐	☐
that feels normal to me	☐	☐	☐	☐	☐
that is typical for me	☐	☐	☐	☐	☐
I find difficult not to do	☐	☐	☐	☐	☐
I will start before I realize that I'm actually doing it	☐	☐	☐	☐	☐

Internet

13) What do you do on the Internet that is related to love and sexuality: (You may tick more than one)

- ☐ I look for someone to have a flirt with
- ☐ I look for a sweetheart/partner
- ☐ I read erotic texts (short stories/stories)
- ☐ I watch erotic pictures/movies
- ☐ I watch pornographic pictures/movies
- ☐ I check out dating sites
- ☐ I respond to sexual contact ads
- ☐ I chat with peers
- ☐ I seek sex education/advice
- ☐ I buy sex products (video, devices, etc.)
- ☐ I contact prostitutes
- ☐ I do nothing related to love or sexuality on the Internet

14) Have you ever met someone on the Internet that you later met off-line and had sex with in real life?

- ○ Yes
- ○ No

If the respondent ticks 'YES' on the question above, question 15-16 will appear on the screen:

15) How many times have you met someone on the Internet that you later had sex with in real life? (enter the number using 2 digits)

16) What was the purpose of meeting the person you met and had sex with in real life (last time it happened?)

- ○ To start a romantic relationship
- ○ To start a sexual relationship
- ○ To have sex only once
- ○ To have sex outside my steady relationship

Substance use

17) Have you ever tried or do you use any of the following substances? (Please, tick one on each line)

	Never tried	Tried	Occasional use	Regular use
Snuff (smokeless tobacco)	☐	☐	☐	☐
Cigarettes	☐	☐	☐	☐
Alcohol	☐	☐	☐	☐
Cannabis (hashish, marijuana)	☐	☐	☐	☐
Amphetamine (speed)	☐	☐	☐	☐
Ecstasy (E)	☐	☐	☐	☐
Other	☐	☐	☐	☐

18) Your feelings about sexuality... (Tick one on each line)

	Strongly agree	Agree	Don't know	Disagree	Strongly disagree
I would really feel nervous if I started a sexual relationship with someone	☐	☐	☐	☐	☐
Sexual fantasies are healthy	☐	☐	☐	☐	☐
I really like the idea of being touched sexually	☐	☐	☐	☐	☐
I really like the idea of me touching someone sexually	☐	☐	☐	☐	☐

19) Do you currently have a girl(boy)friend?

☐ No, I've never had a girl(boy)friend

☐ I had previously, but not now

☐ Yes, I have one now

If the respondent ticks 'YES' on question 19, question 20 will appear on the screen:

20) If you currently have a girl/boyfriend, what gender is she/he?

☐ Boy/male ☐ Girl/female

If the respondent ticks 'I had one previously' on question 19, question 21 will appear on the screen:

21) If you previously had a girl/boyfriend, what gender was she/he?

☐ Gutt/mann ☐ Jente/kvinne

Contraception and chlamydia infections

22) During the past year have you done any of the following?

	Yes	No
Bought condoms?	☐	☐
Received free condoms from the school nurse, the youth clinic or others?	☐	☐
Practiced putting on condom?	☐	☐

23) Have you visited the school nurse, the youth clinic or a medical center to get any of the following during the past 2 years?

	Yes	No
Condoms	☐	☐
Other contraception	☐	☐
Advice/testing because you/your partner suspected pregnancy	☐	☐
Advice for sexually transmitted infections (chlamydia, herpes, HIV, etc.)	☐	☐
Testing or treatment for sexually transmitted infections (chlamydia, herpes, HIV, etc.)	☐	☐

Chlamydia testing and risk-assessment

24) How did you react to being chlamydia tested at school?

	Strongly agree	Agree	Don't know	Disagree	Stronly disagree
I was glad to be offered a test	☐	☐	☐	☐	☐
I reacted negatively to the offer	☐	☐	☐	☐	☐
It did not affect me positively or negatively	☐	☐	☐	☐	☐

25) How do you assess your own risk of being infected by chlamydia?

- ☐ No risk
- ☐ Low risk
- ☐ Medium risk
- ☐ High risk
- ☐ Very high risk
- ☐ Don't know

If the respondent ticks 'No risk' or 'Low risk' on question 25, question 26 will appear on the screen:

26) The reason (-s) why you answered no or low risk is: (You may tick more than one)

- ☐ I never have sex with anyone
- ☐ I have a steady partner
- ☐ I trust my partner to inform about an infection
- ☐ I can assess if my partner has a chlamydia infection
- ☐ I always use a condom
- ☐ Other reasons

Chlamydia infection

27) Have you previously been tested for chlamydia?

- ☐ Yes, once
- ☐ Yes, twice
- ☐ Yes, 3 times
- ☐ Yes, 4 times or more
- ☐ No

28) Have you previously been treated for chlamydia infection?

- ☐ Yes, once
- ☐ Yes, twice
- ☐ Yes, 3 times
- ☐ Yes, 4 times or more
- ☐ No

If the respondent ticks 'Yes, once, twice, 3 times or ≥ 4 times' on question 28, question 29 will appear on the screen:

29) The last time you were treated for chlamydia, was (were) your partner (-s) notified for treatment?

☐ Yes ☐ No ☐ Don't know

If the respondent ticks 'Yes' on question 29, question 30 will appear on the screen:

30) The last time you were treated for chlamydia infection, who notified your partner (-s)?

- ☐ I did it myself
- ☐ The school nurse/public health nurse
- ☐ The physician
- ☐ Other

31) In the past 4 weeks, have you received antibiotic treatment for chlamydia or another infection?

☐ Yes ☐ No ☐ Don't know

If the respondent ticks 'Girl' on Gender, question 32 will appear:

32) Girls: Do you currently have any of the following symptoms? (Place a tick on each line)

	Yes	No
Burning sensation/pain on urination	☐	☐
Changed or increased vaginal discharge	☐	☐
Lower abdominal pain	☐	☐
Vaginal bleeding between periods	☐	☐
Vaginal bleeding after sex	☐	☐

If the respondent ticks 'Boy' on Gender, question 33 will appear:

33) Boys: Do you currently have any of the following symptoms? (Place a tick on each line)

	Yes	No
Pain/burning sensation on urination	☐	☐
Urethral discharge	☐	☐
Testicular swelling or tenderness	☐	☐
Rash/itching/soreness on the penis head	☐	☐

Sexual orientation

34) What is your sexual orientation?

☐ Heterosexual (straight)

☐ Homosexual (gay/lesbian), bisexual

☐ Not sure

35) Have you ever had homosexual experiences?

☐ Yes ☐ No

36) Have you ever had sexual intercourse?

☐ Yes ☐ No

If the respondent ticks 'Girl' on Gender and 'Yes' on question 36, question 37 will appear:

37) Have you ever done any of the following together with a boy?

	Ja	Nei
Hugged and held arms around each other	☐	☐
Kissed	☐	☐
Kissed with tongues	☐	☐
Been touched all over your body by the partner	☐	☐
Touched the partner all over his body	☐	☐

If the respondent ticks 'Boy' on Gender and 'Yes' on question 36, question 38 will appear:

38) Have you ever done any of the following together with a girl?

	Yes	No
Hugged and held arms around each other?	☐	☐
Kissed	☐	☐
Kissed with tongues	☐	☐
Been touched all over your body by the partner	☐	☐
Touched the partner all over her body	☐	☐

If the respondent ticks 'No' on question 36, questions 39-43 will appear:

39) Have you ever wanted to have sexual intercourse?

☐ No ☐ Yes, occasionally ☐ Yes, often

40) Have you ever started a sexual intercourse?

No Yes

41) There may be many reasons for you not having had sexual intercourse. Please highlight the main reasons for not having had sex (You may tick more than one)

- I'm not ready to have sex yet
- I'm too shy
- I'm not interested of sex
- I'm waiting for the right person
- I've got to be in love
- I'll wait until I get married
- I've not had the opportunity
- I think it is wrong/immoral
- My partner does/did not want to
- I'm scared of getting pregnant/my partner getting pregnant
- I'm afraid that my parents would disapporove
- None of my friends have had intercourse yet
- I'm afraid that it will hurt
- Other reasons

42) From the list above, what is the main reason you have not had sexual intercourse yet?

Select answer

43) Please, consider if the following statements about sexuality and sexually transmitted infections are right or wrong.

	Correct	Wrong	Don't know
A woman can get pregnant without the male ejaculating	☐	☐	☐
A woman's chance of getting pregnant increases if she has an orgasm	☐	☐	☐
Menstrual bleeding indicates physical maturation in women	☐	☐	☐
Chlamydia is sexually transmitted and can infect both women and men	☐	☐	☐
Boys cannot have erection before they reach puberty	☐	☐	☐
To get pregnant a women needs to have sex with the male partner more than once	☐	☐	☐
Egg cells origin in the uterus	☐	☐	☐
Masturbation is harmless	☐	☐	☐
'Safe periods' are as safe as birth control pills to prevent pregnancy	☐	☐	☐
Gonorrhea is the most common sexually transmitted infection in Norway	☐	☐	☐

If the respondent ticks 'Yes' on question 36, questions 44-49 will appear:

First sexual intercourse

---

44) How old were you at your first intercourse? (Please, indicate age in two digits, eg. 17)

45) At your first intercourse, how old was your partner? (Please, indicate age in two digits, eg. 17)

46) At your first intercourse, how long had you known your partner?

- ☐ Had met him/her for the first time the same day/evening
- ☐ Less than a week
- ☐ 1-4 weeks
- ☐ 1-6 months
- ☐ More than 6 months

47) Did you use alcohol/drugs in connection with your first intercourse?

- ☐ No
- ☐ Yes, but I was not affected by it
- ☐ Yes, I was a little tipsy
- ☐ Yes, I was very drunk/drugged up

48) There are usually many different reasons for a person to have sexual intercourse. What was your reason to have your first sexual intercourse? (Tick one on each line)

	Yes	No
Because I was in love with my partner	☐	☐
Because my partner wanted to	☐	☐
Because everyone else had done it	☐	☐
Because my partner used pressure	☐	☐
Curiosity/excitement	☐	☐
I was sexually aroused/horny	☐	☐
To gain experience	☐	☐
Don't know, it just turned out that way	☐	☐

49) Did you use any kind of contraception at your first intercourse?

- ☐ No
- ☐ Yes, condoms
- ☐ Hormonal contraception (the pill, p-patch, p-injections, etc)
- ☐ IUD
- ☐ Both condoms and other contraception
- ☐ Emergency pill
- ☐ Other protection
- ☐ Withdrawal (coitus interruptus)
- ☐ Don't know

If the respondent ticks 'Yes' on question 36 and 'No, Hormonal contraception, IUD, Emergency pill, Other, Withdrawal or Don't know' on question 49, question 50 will appear:

50) What were your reasons for not using a condom at first intercourse? (Please tick all reasons that apply)

- ☐ Had no condoms available
- ☐ I was trying for a baby
- ☐ I was not worried of sexually transmitted infections
- ☐ I was unprepared
- ☐ Too drunk/ drugged up
- ☐ I would not risk losing my erection
- ☐ Dared not suggest a condom
- ☐ It feels better without
- ☐ Not sure how to put it on
- ☐ Used other methods of contraception (the pill, IUD, etc)
- ☐ Used other methods of contraception (withdrawal, safe periods)
- ☐ Other

If the respondent ticks 'Yes' on question 36, question 51 will appear:

51) Have you had sexual intercourse more than once?

☐ Yes ☐ No

If the respondent ticks 'Yes' on question 36 and 'Yes' on 51, questions 52-55 will appear:

52) How many sexual partners have you had the last 6 months? (Use two digits, eg. 02)

53) How many sexual partners have you had past 12 months? (Use two digits, eg. 02)

54) What is your lifetime number of sexual partners, ie total number of partners? (Use two digits, eg. 02)

55) How long time has it been since your last sexual intercourse?

☐ Less than a week

☐ 1-4 weeks

☐ 1 month - less than 3 months

☐ 3 months - 1 year

☐ More than 1 year ago

If the respondent ticks 'Yes' on question 36 and 'Less than a week' or '1-4 weeks' on 55, question 56 will appear:

56) Approximately how many times have you had sexual intercourse the past month?

☐ Once

☐ 2-5

☐ 6-9

☐ 10-30

☐ More than 30

If the respondent ticks 'Yes' on question 36, question 57 will appear.

Questions about the *first time* you had intercourse with your *last* sexual partner

---

57) Did you use a condom the first time you had sexual intercourse with your last partner?

☐ Yes ☐ No

If the respondent ticks 'Yes' on question 36 and 'No' on 57, question 58 will appear.

58) If you did not use a condom on that occasion, how well do the following statements agree with your situation just when you had sex? *Do not take too long to think about the answers. (Tick one on each line)*

	Strongly agree	Agree	Don't know	Disagree	Strongly disgaree
I trust my partner	☐	☐	☐	☐	☐
If I pull out a condom, my partner will think that I've been with many others before him/her	☐	☐	☐	☐	☐
If I pull out a condom, my partner will think I have an STI that I will not talk about	☐	☐	☐	☐	☐
If I pull out a condom, I've got no romantic appeal	☐	☐	☐	☐	☐
If I pull out a condom, my partner will think we're together only for the sex	☐	☐	☐	☐	☐
If I pull out a condom, my partner will think that I don't want to go steady	☐	☐	☐	☐	☐
I don't need condoms because I know my partner well	☐	☐	☐	☐	☐
We've got other contraception	☐	☐	☐	☐	☐

If the respondent ticks 'Yes' on question 36, quest. 59-62 will appear

Questions about your *last sexual intercourse*

59) What was your relationship with your last sexual partner?

- ☐ Regular partner/sweetheart
- ☐ Ex-partner/sweetheart
- ☐ A friend I have sex with
- ☐ An acquaintance
- ☐ Casual contact
- ☐ Other

60) At last sexual intercourse, how old was your partner? (Use two digits, eg. 17)

61) How did you meet your last sexual partner, ie. on what occasion?

- ☐ At school/work
- ☐ Through friends/family
- ☐ At a private party
- ☐ At a bar, disco, club
- ☐ On the Internet
- ☐ Other

62) Did you use any contraception at your last intercourse?

- ☐ None
- ☐ Yes, condoms
- ☐ Hormonal contraception (the pill, p-patch, p-injections, etc)
- ☐ IUD
- ☐ Yes, both condoms and other contraception
- ☐ Yes, emergency pill
- ☐ Other protection
- ☐ Withdrawal (coitus interruptus)
- ☐ Don't know

If the respondent ticks 'Yes' on question 36 and 'No, Hormonal contraception, IUD, Emergency pill, Other, Withdrawal or Don't know' on 62, question 63 will appear:

63) Why did you not use condom at last intercourse?

- ☐ Had no condoms available
- ☐ I was trying for a baby
- ☐ Did not worry about sexually transmitted infections
- ☐ I was unprepared
- ☐ Too drunk/ drugged up
- ☐ I would not risk losing my erection
- ☐ Dared not suggest a condom
- ☐ It feels better without
- ☐ Unsure how to put it on
- ☐ Used other contraception (the pill, IUD, etc.)
- ☐ Used other contraception (withdrawal, safe periods)
- ☐ I'm always STI tested after unprotected sex
- ☐ Other

If the respondent ticks 'Yes' on question 36 and 'Yes, condoms' or 'Yes, condoms and other contraception', questions 64-65 will appear:

64) What was (were) your reasons for using a condom at last intercourse? (Please tick all reasons that apply on that occasion)

- ☐ To avoid pregnancy
- ☐ To avoid catching a sexually transmitted infection
- ☐ To avoid HIV/AIDS
- ☐ To be more hygienic
- ☐ To avoid making a mess
- ☐ For fun
- ☐ To make sex last longer
- ☐ To make entry smoother
- ☐ Other reasons

65) From the list above, what was your main reason for using one?

Select answer

If the respondent ticks 'Yes' on question 36, question 66-68 will appear:

66) At your last intercourse, how long had you known your partner?

- Had met him/her for the first time on the same day/evening
- Less than a week
- 1-4 weeks
- 1-6 months
- More than 6 months

67) Did you use alcohol or drugs in connection with last sexual intercourse?

- No
- Yes, but I was not affected by it
- Yes, I was a little tipsy
- Yes, I was very drunk/drugged up

68) What was the reason you had sex the last time? (Please, tick one on each line)

	Yes	No
Because I was in love with my partner		
Because my partner wanted to		
Beacuse my partner used pressure		
Curiosity/excitement		
Was sexually aroused/horny		
Don't know, it just turned out that way		

Appendix E

Questionnaire

Original Norwegian version

Hovedstudie:
Seksualitet og klamydia i Finnmark

Først vil vi vite litt om din bakgrunn og hvordan du ser på deg selv

---

1) * Kjønn

☐ Gutt/mann ☐ Jente/kvinne

2) * Fødselsår, fire siffer (ÅÅÅÅ):

3) * Skolekommune

Velg alternativ

4) Hvor bor du under skolegangen?

☐ Hjemme hos foreldre/andre foresatte

☐ Besteforeldre, andre slektninger

☐ Privat hybel

☐ 'Elevhjem'

☐ Vertsfamilie

☐ Annet

Utdanning

5) Hva er den høyeste utdanningen du har tenkt å ta?

- ☐ Videregående skole: Allmenn, økonomi, administrasjon, idrett eller musikk-dans-drama
- ☐ Videregående skole: Yrkesfag
- ☐ Høyskole eller universitet, 4 år eller mindre (f.eks. bachelor, lærer, politi, sykepleier, ingeniør, journalist)
- ☐ Høyskole eller universitet, mer enn 4 år (f.eks. master, lektor, advokat, sivilingeniør, lege, tannlege)
- ☐ Har ikke bestemt meg
- ☐ Annet

6) Hvilken utdanning har/hadde din mor?

- ☐ Ingen utdanning etter grunnskole
- ☐ Yrkesfaglig videregående skole eller tilsvarende
- ☐ Allmennfaglig videregående skole (gymnas) eller tilsvarende
- ☐ Høyskole eller universitet, 4 år eller mindre (f.eks. bachelor, lærer, politi, sykepleier, ingeniør, journalist)
- ☐ Høyskole eller universitet, mer enn 4 år (f.eks. master, lektor, advokat, sivilingeniør, lege, tannlege)
- ☐ Vet ikke

7) Hvilken utdanning har/hadde din far?

- ☐ Ingen utdanning etter grunnskole
- ☐ Yrkesfaglig videregående skole eller tilsvarende
- ☐ Allmennfaglig videregående skole (gymnas) eller tilsvarende
- ☐ Høyskole eller universitet, 4 år eller mindre (f.eks. bachelor, lærer, politi, sykepleier, ingeniør, journalist)
- ☐ Høyskole eller universitet, mer enn 4 år (f.eks. master, lektor, advokat, sivilingeniør, lege, tannlege)
- ☐ Vet ikke

Kultur og kontakt

8) Jeg oppfatter min etnisitet som: (du kan sette flere kryss)

☐ Norsk ☐ Samisk ☐ Russisk ☐ Kvensk ☐ Finsk ☐ Annet

9) Besteforeldres (mors) etnisitet er: (du kan sette flere kryss)

☐ Norsk ☐ Samisk ☐ Russisk ☐ Kvensk ☐ Finsk ☐ Annet

10) Besteforeldres (fars) etnisitet er: (du kan sette flere kryss)

☐ Norsk ☐ Samisk ☐ Russisk ☐ Kvensk ☐ Finsk ☐ Annet

11) Har du og dine foreldre / foresatte tilhørighet til noe spesielt trossamfunn?

	Statskirke	Læstadianisme	Jehovas vitner, pinsemenighet	Russisk-ortodoks	Islam	Ingen tilhørighet
Meg selv	☐	☐	☐	☐	☐	☐
Mor	☐	☐	☐	☐	☐	☐
Far	☐	☐	☐	☐	☐	☐

Selvbilde

De følgende spørsmålene handler om du opplever negative tanker om deg selv. For hvert utsagn ber vi deg krysse av i boksen som du synes passer best for deg.

12) Å tenke negativt om meg selv er noe...

	Helt enig	Enig	Usikker	Uenig	Helt uenig
jeg gjør ofte	☐	☐	☐	☐	☐
jeg gjør automatisk	☐	☐	☐	☐	☐
som på en måte føles naturlig for meg	☐	☐	☐	☐	☐
som er typisk for meg	☐	☐	☐	☐	☐
jeg har vanskelig for å la være	☐	☐	☐	☐	☐
jeg begynner på før det går opp for meg at jeg gjør det	☐	☐	☐	☐	☐

Internett

13) Hva gjør du på internett som har tilknytning til kjærlighet og seksualitet: (Du kan sette flere kryss)

- ☐ Leter etter noen å flørte med
- ☐ Leter etter kjærlighetskontakter/partnere
- ☐ Leser erotiske tekster (noveller/fortellinger)
- ☐ Ser på erotiske bilder/filmer
- ☐ Ser på pornografiske bilder/filmer
- ☐ Sjekker ut kontaktsider
- ☐ Svarer på sexannonser
- ☐ Chatter med likesinnede
- ☐ Søker seksualopplysning/rådgivning
- ☐ Kjøper sexprodukter (video, hjelpemidler, etc.)
- ☐ Kontakter prostituerte
- ☐ Jeg gjør ingenting knyttet til kjærlighet og seksualitet på internett

14) Har du kommet i kontakt med noen på internett som du etterpå traff og hadde sex med utenfor internett (dvs. i virkeligheten)?

- ○ Ja
- ○ Nei

Hvis respondenten svarer 'ja' på spørsmål 14), vil spørsmål 15-16 komme opp på skjermen.

15) Hvor mange ganger har du kommet i kontakt med noen på internett som du etterpå har hatt sex med utenfor internett? (angi antallet med 2 siffer, f.eks. 02)

_____

16) Hva var formålet med å treffe personen du traff og hadde sex med utenfor internett (den siste gangen det skjedde)?

- ○ For å innlede et romantisk kjærlighetsforhold
- ○ For å innlede et seksuelt forhold
- ○ For å ha sex ved et enkelt tilfelle
- ○ For å ha sex utenfor mitt faste forhold (sidesprang)

Rus

17) Har du noen gang prøvd, eller bruker du, noe av det følgende? (Velg ett alternativ på hver linje)

	Aldri prøvd	Prøvd	Bruker av og til	Bruker regelmessig
Snus	☐	☐	☐	☐
Sigaretter/tobakk	☐	☐	☐	☐
Alkohol	☐	☐	☐	☐
Cannabis (hasj, marihuana)	☐	☐	☐	☐
Amfetamin (speed)	☐	☐	☐	☐
Ecstasy (E)	☐	☐	☐	☐
Annet	☐	☐	☐	☐

18) Dine følelser i forhold til seksualitet (Velg ett alternativ per linje)

	Helt enig	Enig	Usikker	Uenig	Helt uenig
Jeg ville virkelig føle meg nervøs hvis jeg gikk inn i et seksuelt forhold med noen	☐	☐	☐	☐	☐
Seksuelle fantasier er sunt	☐	☐	☐	☐	☐
Jeg liker virkelig tanken på å bli befølt seksuelt	☐	☐	☐	☐	☐
Jeg liker virkelig tanken på å beføle noen seksuelt	☐	☐	☐	☐	☐

19) Har du for tiden en kjæreste?

- ☐ Nei, jeg har aldri hatt en kjæreste
- ☐ Jeg hadde en tidligere, men ikke nå
- ☐ Ja, jeg har en nå

Hvis respondenten svarer 'ja' på spørsmål 19, vil spørsmål 20 komme opp på skjermen.

20) Hvis du har en kjæreste nå, hvilket kjønn har kjæresten din?

☐ Gutt/mann ☐ Jente/kvinne

Hvis respondenten krysser 'Jeg hadde en tidligere, men ikke nå' på spørsmål 19, vil spørsmål 21 komme opp på skjermen.

21) Hvis du hadde en kjæreste tidligere, hvilket kjønn var kjæresten din?

☐ Gutt/mann ☐ Jente/kvinne

Prevensjon og klamydiasmitte

22) Har du i løpet av det siste året gjort noe av det følgende?

	Ja	Nei
Kjøpt kondomer	☐	☐
Fått kondomer gratis fra helsesøster, Ungdommens helsestasjon, organisasjon, el. annet	☐	☐
Øvd på å sette på kondom selv	☐	☐

23) Har du i løpet av de siste 2 årene oppsøkt helsesøster på skolen, Ungdommens helsestasjon eller et legesenter for å få noe av det følgende?

	Ja	Nei
Kondomer	☐	☐
Annen prevensjon	☐	☐
Råd/test i forbindelse med at du selv/partneren mistenkte graviditet	☐	☐
Råd for kjønnssykdommer (klamydia, herpes, hiv, osv.)	☐	☐
Test/behandling for kjønnssykdommer (klamydia, herpes, hiv, osv.)	☐	☐

Reaksjon på test og risiko for klamydiasmitte

24) Reaksjon på å bli klamydiatestet på skolen (ett kryss per linje)

	Helt enig	Enig	Usikker	Uenig	Helt uenig
Jeg er glad for at jeg fikk tilbud om test	☐	☐	☐	☐	☐
Jeg reagerte negativt over å få et slikt tilbud	☐	☐	☐	☐	☐
Tilbud om tester påvirker meg ikke positivt eller negativt	☐	☐	☐	☐	☐

25) Hvor stor risiko bedømmer du at du har for å smittes med klamydia?

- ☐ Ingen risiko
- ☐ Liten risiko
- ☐ Middels risiko
- ☐ Stor risiko
- ☐ Meget stor risiko
- ☐ Usikker/vet ikke

Hvis respondenten krysser 'Ingen risiko' eller 'Liten risiko' på spørsmål 25, vil spørsmål 26 komme opp på skjermen.

26) Årsaken til at du svarte ingen eller liten risiko er: (du kan sette flere kryss)

- ☐ Aldri har sex med noen
- ☐ Har en fast partner
- ☐ Stoler på at partneren forteller om han/hun er smittet
- ☐ Synes du klarer å vurdere på forhånd om partner er smittet eller ikke
- ☐ Alltid bruker kondom
- ☐ Annet

Klamydiainfeksjon

27) Har du tidligere blitt testet for klamydiainfeksjon?

- ☐ Ja, en gang
- ☐ Ja, 2 ganger
- ☐ Ja, 3 ganger
- ☐ Ja, 4 ganger eller flere
- ☐ Nei

28) Har du tidligere fått behandling for en klamydiainfeksjon?

- ☐ Ja, en gang
- ☐ Ja, 2 ganger
- ☐ Ja, 3 ganger
- ☐ Ja, 4 ganger eller flere
- ☐ Nei

Hvis respondenten krysser 'Ja, en, 2, 3, 4 eller flere ganger' på spørsmål 28, vil spørsmål 29 komme opp på skjermen.

29) Siste gang du ble behandlet for klamydia, ble din(-e) partner(-e) kontaktet for behandling?

☐ Ja ☐ Nei ☐ Vet ikke

Hvis respondenten krysser 'Ja' på spørsmål 29, vil spørsmål 30 komme opp på skjermen.

30) Siste gang du ble behandlet for klamydia, hvem tok kontakt med din(-e) partner(-e)?

- ☐ Jeg tok selv kontakt
- ☐ Helsesøster tok kontakt
- ☐ Lege tok kontakt
- ☐ Annet

31) Har du de siste 4 uker blitt behandlet med antibiotika for klamydia eller annen infeksjon?

☐ Ja ☐ Nei ☐ Vet ikke

Hvis respondenten krysser 'Jente' på 'Kjønn', vil spørsmål 32 komme opp:

32) For jenter: Har du for øyeblikket noen av følgende symptomer? (sett ett kryss på hver linje)

	Ja	Nei
Svie ved vannlating	☐	☐
Endret eller økt utflod	☐	☐
Smerter underliv/nedre del av magen	☐	☐
Blødning mellom menstruasjoner	☐	☐
Blødning etter samleie	☐	☐

Hvis respondenten krysser 'Gutt' på 'Kjønn', vil spørsmål 33 komme opp:

33) For gutter: Har du for øyeblikket noen av følgende symptomer? (sett ett kryss på hver linje)

	Ja	Nei
Svie ved vannlating	☐	☐
Utflod fra urinrøret	☐	☐
Øm eller hoven pung	☐	☐
Utslett/kløe/sårhet på penishodet	☐	☐

Seksuell orientering

34) Hva regner du som din seksuelle orientering?

☐ Heterofil/streit

☐ Lesbisk/homofil/bifil/skeiv

☐ Jeg er usikker på min seksuelle orientering

35) Har du hatt sex med en person av samme kjønn som deg selv?

☐ Ja ☐ Nei

36) Har du noen gang hatt samleie?

☐ Ja ☐ Nei

Hvis respondenten krysser 'Jente' på 'Kjønn' og 'ja' på spørsmål 36, vil spørsmål 37 komme opp:

37) Har du sammen med en gutt noen gang:

	Ja	Nei
Gitt han en klem, holdt rundt hverandre	☐	☐
Kysset	☐	☐
Tungekysset	☐	☐
Blitt befølt over hele kroppen av partneren	☐	☐
Befølt partneren over hele kroppen	☐	☐

Hvis respondenten krysser 'Gutt' på 'Kjønn' og 'ja' på spørsmål 36, vil spørsmål 38 komme opp:

38) Har du sammen med en jente noen gang:

	Ja	Nei
Gitt henne en klem, holdt rundt hverandre	☐	☐
Kysset	☐	☐
Tungekysset	☐	☐
Blitt befølt over hele kroppen av partneren	☐	☐
Befølt partneren over hele kroppen	☐	☐

Hvis respondenten krysser 'Nei' på spørsmål 36, vil spørsmål 39-43 komme opp:

39) Har du noen gang hatt lyst til å ha samleie?

☐ Nei ☐ Ja, noen ganger ☐ Ja, ofte

40) Har du noen gang påbegynt et samleie?

Nei Ja

41) Det er mange grunner til at man ikke har hatt samleie. Marker de viktigste grunnene til at du ikke har hatt samleie enda (du kan sette flere kryss)

- Er ikke klar for å ha samleie enda
- Er for sjenert
- Er ikke interessert i sex
- Venter på den rette personen
- Må være forelsket
- Venter til jeg gifter meg
- Har ikke hatt anledning
- Jeg mener det er galt/umoralsk
- Min partner vil/ville ikke
- Redd for å bli gravid/at partneren skal bli gravid
- Redd for at mine foreldre vil mislike det
- Ingen av vennene mine har hatt samleie enda
- Redd for at det skal gjøre vondt
- Annet

42) Fra listen overfor, hva er hovedgrunnen til at du ikke har hatt samleie?

Velg alternativ

43) Vurder om påstandene som følger om seksualitet og kjønnssykdommer er riktig eller gale.

	Riktig	Galt	Vet ikke
En kvinne kan bli gravid under samleiet uten at mannen får utløsning	☐	☐	☐
En kvinnes sjanse for å bli gravid er mye større dersom hun får orgasme under samleie	☐	☐	☐
Menstruasjon er et tegn på kjønnsmodning hos kvinner	☐	☐	☐
Klamydia er en kjønnssykdom som både menn og kvinner kan få	☐	☐	☐
Gutter utvikler ikke evnen til å få ereksjon før de kommer i puberteten	☐	☐	☐
For å bli gravid må en kvinne ha samleie med samme mann flere ganger	☐	☐	☐
Eggceller dannes i livmoren	☐	☐	☐
Onani er ufarlig	☐	☐	☐
Det å satse på "sikre perioder" er like sikkert som bruk av p-piller for å forhindre graviditet	☐	☐	☐
Gonore er den mest utbredte kjønnssykdommen i Norge	☐	☐	☐

Hvis respondenten krysser 'Ja' på spørsmål 36, vil spørsmål 44-49 komme opp:

Første samleie

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44) Ved ditt første samleie: Hvor gammel var du? (angi alder i 2 siffer, f.eks. 17)

45) Ved ditt første samleie: Hvor gammel var din partner? (angi alder i 2 siffer, f.eks. 17)

46) Ved ditt første samleie, hvor lenge hadde du kjent din partner på forhånd?

- ☐ Hadde møtt han/henne for første gang samme dag/kveld
- ☐ Mindre enn 1 uke
- ☐ 1-4 uker
- ☐ 1-6 måneder
- ☐ Over 6 måneder

47) Hadde du drukket alkohol/brukt stoff i forbindelse med ditt første samleie?

- ☐ Nei
- ☐ Ja, men var ikke beruset
- ☐ Ja, og var litt beruset
- ☐ Ja, og var svært beruset

48) Mennesker har samleie av mange grunner. Hva var grunnen til at du hadde samleie akkurat første gangen? (sett ett kryss på hver linje)

	Ja	Nei
Fordi jeg var forelsket/glad i partneren min	☐	☐
Fordi partneren ville det	☐	☐
Fordi alle andre hadde hatt det	☐	☐
Fordi jeg ble presset til det	☐	☐
Nysgjerrighet/spenning	☐	☐
Var seksuelt opphisset/kåt	☐	☐
For å få erfaringen	☐	☐
Vet ikke, bare ble slik	☐	◉

49) Brukte dere noen form for beskyttelse ved ditt første samleie?

- [ ] Nei, ingen
- [ ] Ja, kondom
- [ ] Ja, p-piller, p-stav, p-ring, p-plaster, p-sprøyte
- [ ] Ja, spiral
- [ ] Ja, både kondom og annet prevensjonsmiddel
- [ ] Ja, angrepille/dagen-derpå-pille/nødprevensjon
- [ ] Ja, annen beskyttelse
- [ ] Avbrutt samleie
- [ ] Usikker/vet ikke

Hvis respondenten krysser 'Ja' på spørsmål 36, og 'Nei ingen', 'p-piller osv', 'Spiral', 'Angrepille', 'Annen beskyttelse', 'Avbrutt samleie', 'Usikker' på spørsmål 49, vil spørsmål 50 komme opp:

50) Hva var årsaken(-e) til at du ikke brukte kondom ved ditt første samleie? (du kan sette flere kryss)

- [ ] Hadde ingen kondom for hånden
- [ ] Jeg ville ha barn
- [ ] Var ikke urolig for å smittes av kjønnssykdommer
- [ ] Var uforberedt på situasjonen
- [ ] Var påvirket av alkohol/narkotika
- [ ] Ville ikke risikere å miste ereksjonen
- [ ] Våget ikke foreslå kondom
- [ ] Det er deiligere uten
- [ ] Usikker på hvordan man setter på en kondom
- [ ] Brukte annet prevensjonsmiddel(p-piller, spiral, etc.)
- [ ] Brukte annen prevensjonsmetode (avbrutt samleie eller såkalte sikre perioder)
- [ ] Annet

Hvis respondenten krysser 'Ja' på spørsmål 36, vil spørsmål 51 komme opp:

51) Har du hatt mer enn ett samleie?

☐ Ja ☐ Nei

Hvis respondenten krysser 'Ja' på spørsmål 36 og 'Ja' på spørsmål 51, vil spørsmål 52-55 komme opp:

52) Hvor mange samleiepartnere har du hatt i løpet av de siste 6 månedene? (angi hele tall i to siffer, f.eks. 02)

53) Hvor mange samleiepartnere har du hatt i løpet av de siste 12 månedene? (angi hele tall i to siffer, f.eks. 02)

54) Hvor mange samleiepartnere har du hatt totalt? (angi hele tall i to siffer, f.eks. 02)

55) Hvor lenge er det siden siste gang du hadde samleie?

☐ Mindre enn 1 uke

☐ 1-4 uker

☐ 1 måned-mindre enn 3 måneder

☐ 3 måneder til 1 år

☐ Over 1 år siden

Hvis respondenten krysser 'Ja' på spørsmål 36 og 'Mindre enn 1 uke' eller '1-4 uker' på spørsmål 55, vil spørsmål 56 komme opp:

56) Omtrent hvor mange samleier har du hatt den siste måneden?

☐ 1 samleie

☐ 2-5 samleier

☐ 6-9 samleier

☐ 10-30 samleier

☐ Over 30 samleier

Hvis respondenten krysser 'Ja' på spørsmål 36, vil spørsmål 57 komme opp:

Spørsmål om *den første gangen* du hadde samleie med din *siste* sex-partner

57) Ble det brukt kondom første gang du hadde samleie med din siste partner?

☐ Ja ☐ Nei

Hvis respondenten krysser 'Ja' på spørsmål 36 og 'Nei' på 57, vil spørsmål 58 komme opp:

58) Hvis du ikke brukte kondom ved denne anledningen, hvor godt stemmer påstandene nedenfor med din situasjon akkurat da du hadde samleie

Ikke bruk for lang tid til å tenke gjennom svarene (Sett ett kryss per linje)

	Stemmer svært godt	Stemmer ganske godt	Usikker	Stemmer ganske dårlig	Stemmer svært dårlig
Jeg stoler på partneren	☐	☐	☐	☐	☐
Om jeg drar opp et kondom, vil partneren tro jeg har vært sammen med mange før han/henne	☐	☐	☐	☐	☐
Om jeg drar opp et kondom, vil partneren tro jeg har en kjønnssykdom som jeg ikke vil fortelle om	☐	☐	☐	☐	☐
Om jeg drar opp et kondom, vil det bli uromantisk	☐	☐	☐	☐	☐
Om jeg drar opp ett kondom, vil partneren tro jeg bare er ute etter sex	☐	☐	☐	☐	☐
Om jeg drar opp et kondom, vil partneren tro jeg ikke er ute etter å bli sammen med han/henne	☐	☐	☐	☐	☐
Kondom er unødvendig fordi jeg kjenner partneren godt fra før	☐	☐	☐	☐	☐
Vi har annen prevensjon	☐	☐	☐	☐	☐

Hvis respondenten krysser 'Ja' på spørsmål 36, vil spørsmål 59-62 komme opp:

Spørsmål om *siste* gang du hadde samleie

59) Hvilket forhold har du til den du hadde samleie med siste gang?

- ☐ Fast partner/kjæreste
- ☐ Tidligere partner/kjæreste
- ☐ Venn ("knullevenn/knullevenninne")
- ☐ Bekjent
- ☐ Tilfeldig kontakt/en jeg ikke kjente på forhånd
- ☐ Annen

60) Ved ditt siste samleie: Hvor gammel var din partner? (angi alder i 2 siffer, f.eks. 17)

61) Hvor traff du din siste partner? Ved hvilken anledning?

- ☐ Gjennom skole/arbeid
- ☐ Gjennom venner/familie
- ☐ På privatfest
- ☐ På et utested (diskotek, kafe, bar, osv.)
- ☐ På Internett
- ☐ Annet

62) Brukte dere noen form for beskyttelse ved ditt siste samleie?

- ☉ Nei, ingen
- ☐ Ja, kondom
- ☐ Ja, p-piller, p-stav, p-ring, p-plaster, p-sprøyte
- ☐ Ja, spiral
- ☐ Ja, både kondom og annet prevensjonsmiddel
- ☐ Ja, angrepille/dagen-derpå-pille/nødprevensjon
- ☐ Ja, annen beskyttelse
- ☐ Avbrutt samleie
- ☐ Usikker/vet ikke

Hvis respondenten krysser 'Ja' på spørsmål 36, og på spørsmål 62: 'Ingen, p-piller osv, Spiral, Angrepille, Annen, Avbrutt eller Vet ikke', vil spørsmål 63 komme opp:

63) Hvorfor brukte du ikke kondom ved ditt siste samleie? (du kan sette flere kryss)

- ☐ Hadde ingen kondom for hånden
- ☐ Jeg ville ha barn/var gravid
- ☐ Var ikke urolig for å smittes av kjønnssykdom
- ☐ Var uforberedt på situasjonen
- ☐ Var påvirket av alkohol/narkotika
- ☐ Ville ikke risikere å miste ereksjonen
- ☐ Våget ikke foreslå kondom
- ☐ Det er deiligere uten
- ☐ Usikker på hvordan man setter på et kondom
- ☐ Brukte annet prevensjonsmiddel (p-piller, spiral, etc.)
- ☐ Brukte annen prevensjonsmetode (avbrutt samleie eller såkalte sikre perioder)
- ☐ Tar alltid en test for kjønnssykdom etter ubeskyttet sex
- ☐ Annet

Hvis respondenten krysser 'Ja' på spørsmål 36, og 'Kondom' eller 'Kondom og annen prevensjon' på spørsmål 62,vil spørsmål 64-65 komme opp:

64) Hvilke grunner hadde du for å bruke kondom ved ditt siste samleie? (du kan sette flere kryss)

- ☐ For å unngå graviditet
- ☐ For å unngå smitte med kjønnssykdom
- ☐ For å unngå smitte med HIV/AIDS
- ☐ For å være mer renslig
- ☐ For å unngå å søle
- ☐ For moro skyld
- ☐ For å få sexen til å vare lenger/ikke komme så fort
- ☐ For å gjøre inntrenging lettere
- ☐ Annet

65) Fra listen overfor, hva var hovedgrunnen til kondombruken?

Velg alternativ

Hvis respondenten krysser 'Ja' på spørsmål 36, vil spørsmål 66-68 komme opp:

66) Ved ditt siste samleie, hvor lenge hadde du kjent din partner på forhånd?

- ◉ Hadde møtt han/henne for første gang samme dag/kveld
- ☐ Mindre enn 1 uke
- ☐ 1-4 uker
- ☐ 1-6 måneder
- ☐ Over 6 måneder

67) Hadde du drukket alkohol/brukt stoff i forbindelse med ditt siste samleie?

- ◉ Nei
- ☐ Ja, men var ikke beruset
- ☐ Ja, og var litt beruset
- ☐ Ja, og var svært beruset

68) Hva var grunnen til at du hadde samleie akkurat siste gangen? (Sett ett kryss på hver linje)

	Ja	Nei
Fordi jeg var forelsket/glad i partneren min	☐	☐
Fordi partneren ville det	☐	☐
Fordi jeg ble presset til det	☐	☐
Nysgjerrighet/spenning	☐	☐
Var seksuelt opphisset	☐	☐
Vet ikke, ble bare slik	☐	☐

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