Results Prior to *N gonorrhoeae* inoculation, mice with a pre-existing chlamydial infection had decreased expression of TLR4 and antimicrobial peptide (CRAMP, SLPI) genes. Consistent with the finding of decreased TLR4expression in coinfected mice, markers of inflammation (TLR2, TNFα, IL-1ß, platelet activating factor receptor [Pafr], and IL-23α) were up-regulated only in mice infected with *N gonorrhoeae* alone. A significantly lower percentage of TLR4-expressing epithelial cells was detected in vaginal swabs from chlamydia-infected wild-type mice prior to gonococcal challenge, and importantly, chlamydial infection did not enhance *N gonorrhoeae* infection of TLR4 mutant mice.
Conclusions These data suggest the host response to chlamydial infection creates an environment that is less protective against gonococcal infection by down-regulating the expression of TLR4 and antimicrobial peptides. This work therefore further illuminates the basis of this interesting consequence of coinfection and may also help direct the development of immunomodulatory therapies against this common coinfection and its consequences on reproductive health.

O4-S2.02 SEROPREVALENCE OF NOVEL IMMUNOGENS OF CHLAMYDIA TRACHOMATIS AND THEIR CYTOKINE RESPONSE IN PBMC CELLS UNDER IN VITRO CONDITIONS

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Background Chlamydia trachomatis (CT) is an obligate intracellular parasite which causes STD and trachoma. Despite major research into Chlamydial pathogenesis and host immune responses, immuno-protection has been hampered by the incomplete understanding of protective immunity in the genital tract. Characterised vaccine candidates in general have shown variable efficacy ranging from no protection to partial protection. It is therefore a research priority to identify novel Chlamydial antigens that may elicit protective immune responses.
Objectives The goal of the present study was to assess the seroprevalence to pkn1and DNA j following natural CT infection in human. The prospects of pkn1 as a Type third secretion substrate and DNA j a non surface Chlamydial protein as potential antigen, prompted us to explore the immunogenic potential of both protein.
Methods pkn, DNA j and ompA were cloned in bacterial expression vector pTrcHis. Ni+-NTA affinity chromatography was used to purify the recombinant proteins. Antigenic stretches of Pkn1, DNA j and OmpA were identified using Bcepred web server, designed for identification of subunit vaccine candidate by Bioinformatics Centre of IMTECH Chandigarh, India. To validate the bioinformatics based analysis, sera of human patient were used to determine serorecactivity of pkn1 and DNA j proteins. OmpA was used as a positive control during the study.
Results Present study showed a high seroprevalence of antibodies against Pkn1 and OmpA ($p<0.001$) in sera of humans infected with CT. while, no antibodies were observed for DNA j. Our studies have shown an association between release of TNF-α and IFN-y levels upon stimulation of PBMC with Pkn1 and OmpA. Cytokine expression profiling (IL-1ß, TNF-α, IL-2 , IL10 and IFN-y) of Human PBMCs in response to Pkn1 stimulation demonstrate for the first time that Pkn1 is a novel immunodominant Chlamydial antigen that is capable of influencing both Th1 and Th2 immune responses by stimulating the release of both Humoural and Cell-mediated regulatory cytokines.
Conclusions Our study demonstrated strong serological responses to Pkn1 and major outer membrane in natural human infection suggesting the role of pkn1 in immune response. Studies are in progress to check how the immunomodulation by Pkn1 alters host-pathogen interactions.

O4-S2.03 AN ANTI-ADHESIVE APPROACH TO PREVENTION OF *C TRACHOMATIS* INFECTION

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Background We have reported that the major outer membrane of *C trachomatis* is glycosylated and the glycan is a high mannose oligosaccharide. Cumulative studies have demonstrated that the glycan is important in attachment and infectivity through binding to the mannose receptor (MR). Glycan removal decreases infectivity in vitro and in vivo and simultaneous administration of mannan, a ligand of the MR, abrogates *C trachomatis* infection in a mouse model of pneumonitis. Thus, we are investigating the feasibility of an anti-adhesive therapy to prevent *C trachomatis* infection by identifying oligosaccharides that are effective in inhibiting infection in vitro and testing their efficacy in a mouse model of genital tract infection.
Methods HeLa 229 cell monolayers were pretreated with serial concentrations of oligosaccharides prior to infection with *C trachomatis*. Neutralisation of infectivity was scored as >50% inhibition. For animal experiments, 8-week-old Swiss Webster female mice were primed with subcutaneous injections of Depo-Provera 1 week prior to challenge. Subsequently, mice were inoculated intravaginally with carbohydrates or PBS (n=5 mice per group) 30 min prior to infection with *C trachomatis*. Vaginal swab samples were obtained at 24, 48, and 72 h. post-infection, at peak times of shedding. Statistical significance was determined by the Student's t test.
Results Carbohydrates have been tested in vitro in hapten inhibition experiments against three serovars (D, E, F) most frequently isolated from genital tract infection. At the highest concentration tested, 4-nitrophenyl-α-D-mannopyranoside inhibited infectivity by 91%–92%; α-D-mannose-PAA from 77 to 93%; hen ovalbumin by 85%–89%; mannan by 77%–83%; and the high mannose fraction prepared from ovalbumin by 59%–98%. To determine efficacy in vivo, Swiss Webster mice were inoculated with different concentrations of inhibitors. Of those tested thus far, shedding of organism was significantly decreased ($p<0.05$). The maximum inhibitions observed were: 4-nitrophenyl-α-D-mannopyranoside (86%), α-D-mannose-PAA (81%), mannan (93%), and the high mannose fraction from ovalbumin (94%).
Conclusions These preliminary studies suggest the potential feasibility for developing an "anti-adhesive therapy" as an alternate topical microbicide approach to prevent *C trachomatis* genital tract infection.

O4-S2.04 CAN A CERVICAL BARRIER PREVENT CHLAMYDIA INFECTION IN THE PIGTAILED MACAQUE CERVICAL CHALLENGE MODEL?

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Background Numerous microbicides that inactivate *Chlamydia trachomatis* in vitro have failed to prevent transmission of this pathogen in the pigtailed macaque cervical challenge model. Since *C trachomatis* replicates in endocervical columnar epithelium but not in the squamous vaginal epithelium, we tested whether a cervical barrier would improve protection when used in conjunction with an otherwise non-protective microbicide.

LETTER

Unusual increase in reported HIV/AIDS cases among older persons in western Hunan province, China

An unusual increase in HIV/AIDS cases among older people was reported to the Hunan Centers for Disease Control between 2005 and 2007. Cases originated in four rural, western districts of this inland province of China. Given the historical concern for outbreaks of HIV in rural areas due to blood donation,[1] these cases prompted closer examination to understand the reasons for their appearance and to take measures to prevent further spread.

Eighty cases met our investigation criterion of 50 years or older and underwent a structured interview. The median age was 65 years (range 51–82); 42% were female. Most were ethnic minorities, 76% Tujia and 9% Miao, with low education. Nearly all had been married; 43% were widowed. Most (54%) spent time away from their spouse (median >5 years); 10% were currently sexually active with a spouse; few ever used condoms with their spouse.

Investigation of the possible modes of HIV acquisition suggests most infections among men were from female sex workers (83% paid for sex, two-thirds in the last 5 years), and among women through infected husbands. One case had a history of selling blood, most recently in 1981. Eight received a blood transfusion, three before 1976 and five after 1984. All denied drug use; all men denied male–male sex; all women denied extramarital sex. Among men buying sex, 97% never used condoms.

Two-thirds had never heard of HIV prior to their diagnosis. Few (14%) knew HIV could be transmitted sexually, through blood (11%) or from mother to child (4%). Of the men reporting commercial sex contact, 82% had no knowledge that condoms could prevent HIV. Most cases (86%) were detected incidentally during the course of treatment for other diseases or because their spouse was HIV-positive. By interview, 78% indicated their spouse had tested for HIV, of whom 69% were reported to be positive.

Our investigation highlights that basic information on HIV/AIDS is not reaching all parts of China, and may especially lag among rural and older people. Discussion of sex with older people has been taboo in China, presenting special challenges in finding effective ways to reach them. As treatment extends survival, the cohort of persons living with HIV will also age. The movement of people between urban and rural areas, an ageing population and the shift of the HIV/AIDS epidemic to sexual transmission[2] are three trends in China that may now have a dangerous intersection.

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Competing interests None.

Patient consent Obtained.

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Contributors XC oversaw and coordinated the fieldwork. JZ, JMH and BYQ conducted the fieldwork. YH completed the survey design, data analysis and drafting of the manuscript. LW and NW provided technical support during and prior to the survey and mobilised some funds to complete the survey.

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