Community-based mpox and sexually transmitted disease surveillance using discarded condoms in the global south

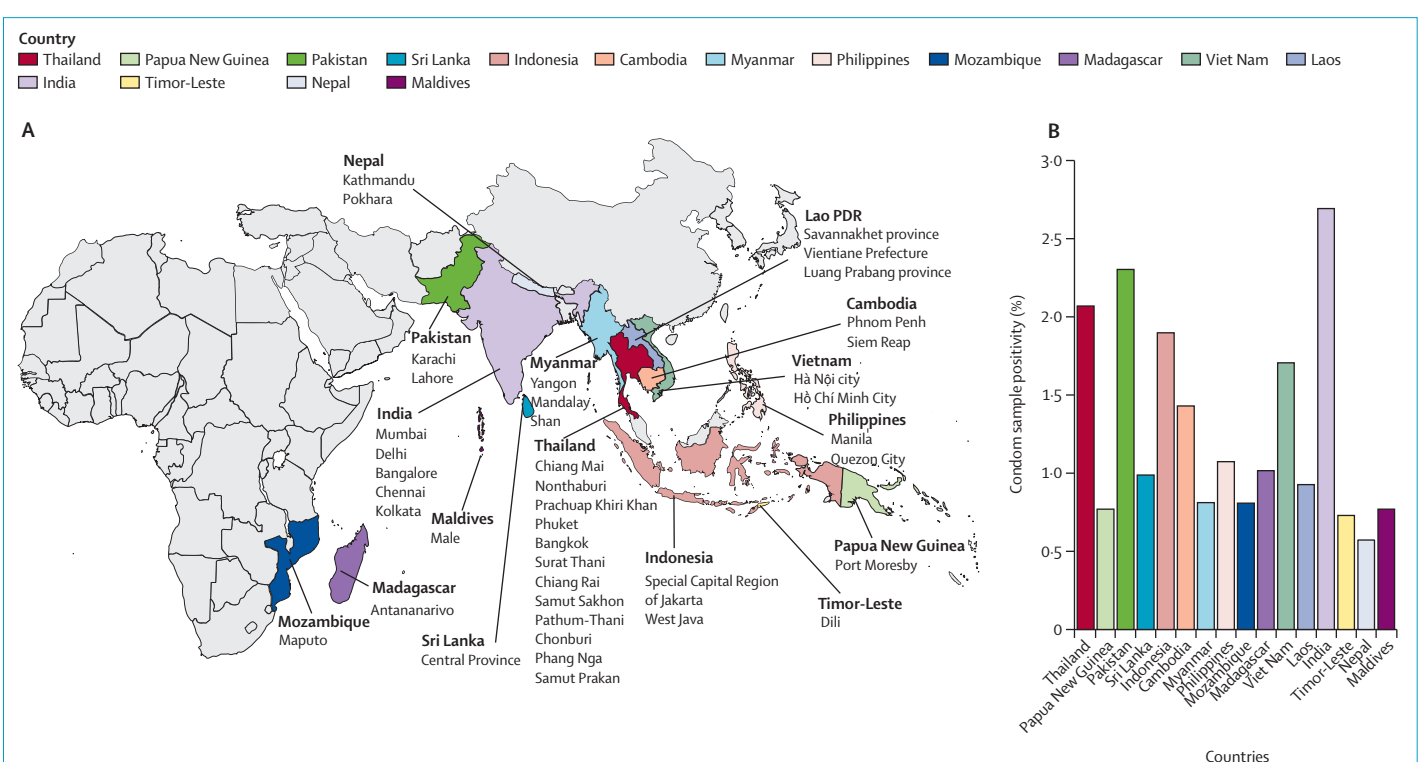
As COVID-19 restrictions have eased, the resumption of international travel and sexual interactions at large gatherings have fueled the continued spread of human mpox beyond its endemic regions.[1–3] The current global circulation of the clade II variant of the monkeypox virus (MPXV) has been marked by unique person-to-person transmission, including sexual and non-sexual contact.[1,3,4] The current transmission dynamics of MPXV indicate changes in the virus's biological characteristics, human behavior, and clinical presentations, making surveillance particularly challenging in resource-limited countries.

To address the surveillance challenges we implemented a community-based surveillance system for mpox using used and discarded condoms. There were 20 941 condoms collected from 16 countries (Thailand, Pakistan, Sri Lanka, Indonesia, Cambodia, Myanmar, the Philippines, Papua New Guinea, Mozambique, Madagascar, Viet Nam, Laos, India, Timor-Leste, Nepal, and the Maldives) with the support of scavengers (members of the public) and sanitary workers. The condoms were collected in a range of places from brothels to public spaces (figure A). MPXV DNA concentrations were quantified using real-time PCR, and the *N3R/OPG016* gene was Sanger-sequenced to validate the real-time PCR results.[2,5] The co-occurrence of other sexually transmitted diseases in MPXV-positive condom samples was also quantified using real-time PCR (appendix).

We found that 262 (1·3%) of 20 941 collected condom samples contained MPXV DNA, with India having the highest positivity rate at 32 (2·7%) of 1188, followed by Pakistan (30/1302 [2·3%]), and Thailand (26/1256 [2·1%]). The lowest positivity rates were observed in Nepal (8/1397 [0·6%]), Papua New Guinea (11/1429 [0·8%]), and the Maldives (11/1429 [0·8%]; figure B). MPXV DNA concentrations in condoms varied spatially and temporally among different countries, ranging from 58·71 to 374·53 copies/mL (figure C). All MPXV DNA in condom samples belonged to the clade IIb (previously called the west African clade; figure D). Sequences from Thailand, Cambodia, the Philippines, Laos, and Indonesia shared the nucleotide substitution C190772A, leading to the amino acid change D121E. Sequences from Papua New Guinea, the Philippines, Viet Nam, Laos, Timor-Leste, Indonesia, and the Maldives exhibited G190660A, causing the amino acid alteration R84K. Samples from Thailand and Nepal showed the substitution C190834T, resulting in

Lancet Infect Dis 2024
Published Online
August 21, 2024
https://doi.org/10.1016/S1473-3099(24)00514-0

See Online for appendix



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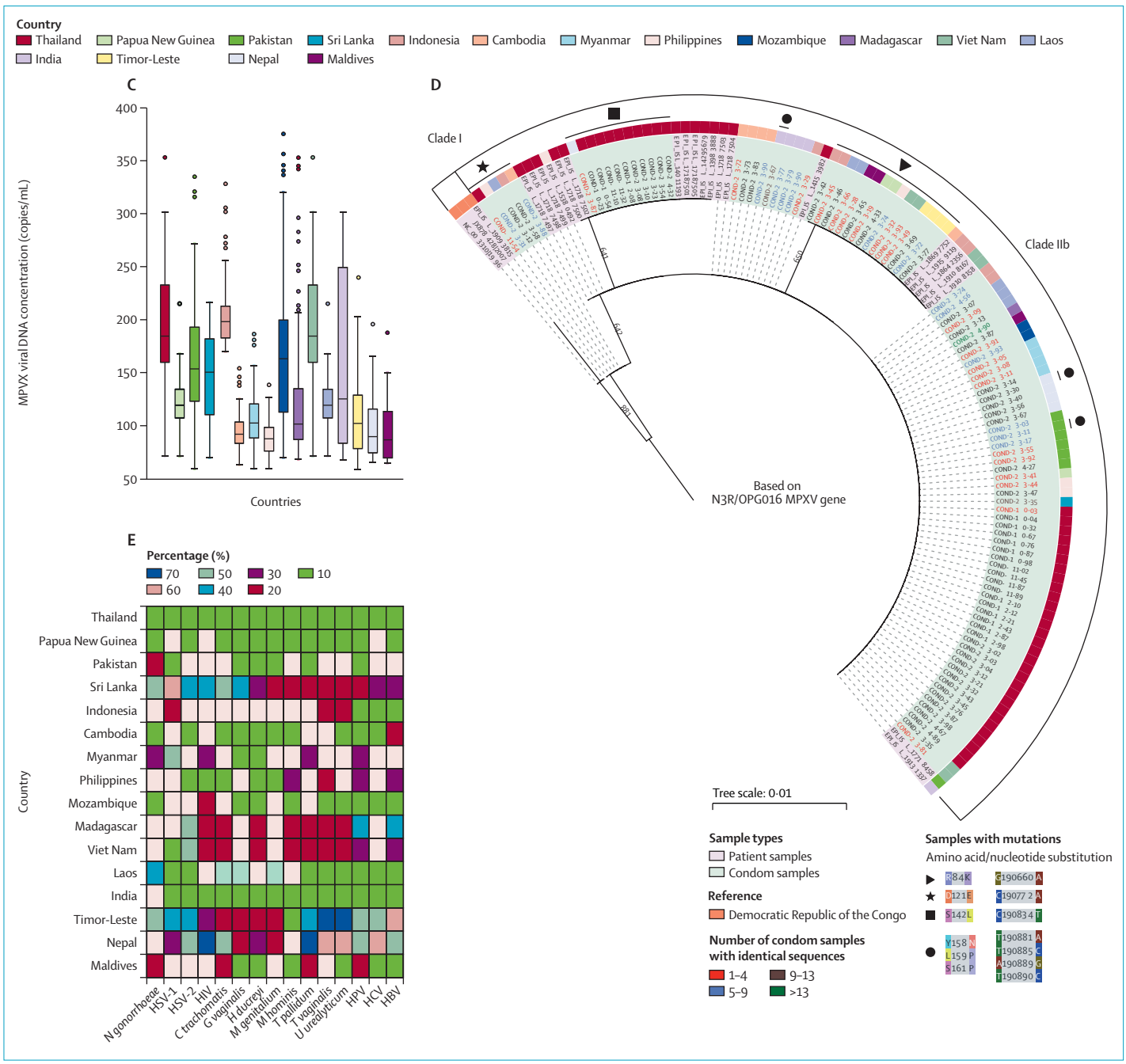

Figure: **Mpox surveillance using discarded condoms in the global south**
(A) Map of condom sampling countries (Thailand [n=1256], Papua New Guinea [n=1429], Pakistan [n=1302], Sri Lanka [n=1315], Indonesia [n=1264], Cambodia [n=1188], Myanmar [n=1233], Philippines [n=1397], Mozambique [n=1485], Madagascar [n=1377], Viet Nam [n=938], Laos [n=1510], India [n=1188], Timor-Leste [n=1233], Nepal [n=1397], Maldives [n=1429]). (B) Condom sample positivity rates among the sampling countries. (C) MPXV DNA concentrations (copies/mL) in positive condom samples. (D) Phylogenetic analysis of MPXV sequences derived from condom samples and GISAID sequences. Evolutionary history was inferred using the Maximum Likelihood method and the Hasegawa–Kishino–Yano substitution model for the N3R/OPG016 gene. The trees with the highest log likelihood are shown. Nextclade (https://clades.nextstrain.org) were used for clade assignment, mutation calling, and quality control for viral genomes. (E) Co-occurrence of sexually transmitted diseases in MPXV-positive condom samples for each country, expressed as a percentage of total MPXV-positive samples. Co-occurrence of sexually transmitted diseases in MPXV-positive condom samples for each country, expressed as a percentage of total MPXV-positive samples. HBV=hepatitis B virus. HCV=hepatitis C virus. HPV=human papillomavirus. HSV=herpes simplex virus. MPXV=monkeypox virus.

S142L. Sequences from India, Nepal, and Pakistan shared four nucleotide substitutions (T190881A, T190885C, A190889G, and T190890C), resulting in the amino acid changes (Y158N, L159P, and S161P; figure D). Samples from the same or neighbouring countries were often grouped closely, suggesting regional outbreaks.

Samples from India, Nepal, and Pakistan showed regional linkage, as did those from Cambodia, Indonesia, and Myanmar. Samples from the Philippines and Viet Nam were closely related. There was a co-occurrence of MPXV-positive condoms with some of the sexually transmitted diseases (*Neisseria gonorrhoeae*, herpes simplex virus type 1 [HSV-1], herpes simplex virus type 2 [HSV-2], HIV, *Chlamydia trachomatis, Gardnerella vaginalis, Haemophilus ducreyi, Mycoplasma genitalium, Mycoplasma hominis, Treponema pallidum, Trichomonas vaginalis, Ureaplasma urealyticum*, human papillomavirus [HPV], hepatitis C virus [HCV], hepatitis B virus [HBV]) analysed (figure E). For *N gonorrhoeae*, the highest co-occurrence was in Sri Lanka (7/13 [53·8%] of the MPXV positive condoms) and Timor-Leste (5/9 [55·6%]). HSV-1 was most commonly co-occurring in Sri Lanka (8/13 [61·5%]) and Myanmar (5/10 [50·0%]). HSV-2 had the highest co-occurrence in Madagascar (8/14 [57·1%]) and Viet Nam (8/16 [50·0%]). HIV was most prevalent in Nepal (6/8 [75·0%]) and Timor-Leste (3/9 [33·3%]). *C trachomatis* had the highest co-occurrence in Sri Lanka (7/13 [53·8%]) and Pakistan (4/30 [13·3%]). HPV was notably high in Timor-Leste (5/9 [55·6%]) and Madagascar (6/14 [42·9%]). HCV was highest in Papua New Guinea (2/11 [18·2%]). HBV had high co-occurrence in Timor-Leste (6/9 [66·7%]) and Nepal (4/8 [50·0%]).

To our knowledge, our findings demonstrate for the first time the usefulness of used condoms as an indirect surveillance tool for mpox transmission and community mpox co-occurrence with other sexually transmitted diseases. The routes to infection are numerous including MPXV in semen, genital lesions on the penis, vagina, or rectum, and potential contamination from the mouth or throat during oral sex.[1,4,5] There is also a potential risk of co-transmission of mpox with other sexually transmitted diseases, especially HSV, HIV, HCV, and gonorrhoea. It is difficult to know whether the condoms acted as a barrier to the spread or not. Nevertheless, the findings demonstrate that used condoms can be used as an indirect surveillance tool for mpox transmission and community mpox co-occurrence with other sexually transmitted diseases. The current spread of MPXV clade II has a relatively low death rate;[3] however, young children and immunocompromised individuals, including people with HIV, are at increased risk for severe outcomes.[1,3] Furthermore, MPXV has disproportionately affected men who have sex with men, indicating amplified transmission through sexual networks. This underscores the importance of considering multiple transmission routes in mpox surveillance and prevention.

DLW was supported by Balvi Filantropic Fund, Barry Satz and Ronald Satz Memorial Fund and Chulalongkorn University (Second Century Fund-C2F Postdoctoral Fellowship), University of Western Australia (Overseas Research Experience Fellowship) and Yamagata Prefectural Central Hospital, Yamagata, Japan (Clinical Residency Fellowship). CM was supported by the Centre of Excellence in Mathematics, Ministry of Higher Education, Science, Research and Innovation, Thailand, Center of Excellence on Medical Biotechnology, and Thailand Center of Excellence in Physics. AKi is a Rothwell Family Fellow. The funder(s) had no role in study design; in the collection, analysis, and interpretation of data; in the writing of the report; or in the decision to submit the article for publication. All other authors declare no competing interests. The data supporting the findings of this study are available within this paper and its additional information. All genome sequences and associated metadata in this study are published in GISAID's EpiCoV database under the mpox category (accession numbers are given in the appendix). The institutional review board at Yamagata Prefectural Central Hospital, Yamagata, Japan, waived ethical approval for this work as it determined that our project is exempt under the type of environmental surveillance study. We, the authors of this paper, embrace inclusive, diverse, and equitable conduct of research. Our team comprises individuals who self-identify as underrepresented ethnic minorities, gender minorities, members of the LGBTQIA+ community, and individuals living with disabilities. We actively promote gender balance in our reference list while maintaining scientific relevance. We thank all the volunteers, entertainment venue staff, and sanitary workers who kindly supported the sample collection. Also, thanks to the United States Centers for Disease Control and Prevention (CDC) Thailand for technical support and the previous Chargé d'Affaires of the United States of America to Thailand (U.S. Embassy & Consulate in Thailand) for facilitating collaboration with the CDC and Armed Forces Research Institute of Medical Sciences. We also thank the LGBTQIA+ community in Indonesia, Madagascar, Myanmar, Thailand, Papua New Guinea, Mozambique, Pakistan, Viet Nam, Sri Lanka, India, Philippines, Cambodia, Laos, Timor-Leste, Nepal, Maldives for generously supporting sample collection, TEDxChiangMai team for facilitating a platform for collaboration, and marginalised, vulnerable indigenous communities in Indonesia, Madagascar, Myanmar, Thailand, Papua New Guinea, Mozambique, Pakistan, Viet Nam, Sri Lanka, India, the Philippines, Cambodia, Laos, Timor-Leste, Nepal, and the Maldives for supporting sample collection. Special thanks to Nuttawut Kietchaiyakorn for helping with the illustrations.

Editorial note: The Lancet Group takes a neutral position with respect to territorial claims in published maps.

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1 Mitjà O, Alemany A, Marks M, et al. Mpox in people with advanced HIV infection: a global case series. *Lancet* 2023; **401:** 939–49.

2 Wannigama DL, Amarasiri M, Phattharapornjaroen P, et al. Tracing the transmission of mpox through wastewater surveillance in Southeast Asia. *J Travel Med* 2023; **30:** taad096.

3 Thornhill JP, Barkati S, Walmsley S, et al. Monkeypox virus infection in humans across 16 countries — April–June 2022. *N Engl J Med* 2022; **387:** 679–91.

4 Lapa D, Carletti F, Mazzotta V, et al. Monkeypox virus isolation from a semen sample collected in the early phase of infection in a patient with prolonged seminal viral shedding. *Lancet Infect Dis* 2022; **22:** 1267–69.

5 Wannigama DL, Amarasiri M, Hongsing P, et al. Multiple traces of monkeypox detected in non-sewered wastewater with sparse sampling from a densely populated metropolitan area in Asia. *Sci Total Environ* 2023; **858:** 159816.